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Back numbers, £10 (U.S.A. $28.00) per volume

Single issues, £2. 10s. (U.S.A. $7.00) each

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Geochimica et Cosmochimica Acta, 1960, Vol. 19, p. 235. Pergamon Press Ltd. Printed in Northern Ireland

Symposium on the Geochemistry of Petroleum

FOREWORD

THE papers published in the present issue of *Geochimica et Cosmochimica Acta* formed a part of The General Petroleum Geochemistry Symposium, which was held on 4 June 1959, during the Fifth World Petroleum Congress, at Fordham University in New York. Summaries of these and other papers were preprinted prior to the Symposium and they were discussed from the floor by the participants who came from approximately thirty different countries.

Petroleum geochemistry and the larger field of organic geochemistry encompass a variety of disciplines. Bacteriology, soil science, organic chemistry, oceanography, clay mineralogy, etc., are the branches of science with which organic geochemistry is associated and through which organic geochemists interpret their observations. It is hoped that the papers in this issue of *Geochimica et Cosmochimica Acta* may further stimulate interdisciplinary exchange of ideas.

The symposium itself stimulated such an exchange of ideas to the extent that immediately following the meeting the suggestion was made that a group on organic geochemistry be organized within the Geochemical Society. The Society approved the idea at its meeting in November 1959, and the group was formally organized at that time. Any member of the Geochemical Society who wishes to do so can affiliate with this group by indicating his interest to the Secretary of the Society, or to the undersigned, who is its temporary chairman.

It is a pleasure to record here the hard work and the invaluable assistance in getting the Proceedings of the Symposium ready for publication rendered by the special associate editors, WARREN G. MEINSCHEIN and IRVING A. BREGER.

28 *January* 1960

BARTHOLOMEW NAGY
Special Editor of the Symposium
on the Geochemistry of Petroleum

Geochimica et Cosmochimica Acta, 1960, Vol. 19, pp. 236 to 243. Pergamon Press Ltd. Printed in Northern Ireland

Recovery of dissolved organic matter in sea-water and organic sorption by particulate material*

R. G. Bader, D. W. Hood and J. B. Smith
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College Station, Texas

Abstract—A brief discussion on some initial data concerning organic matter dissolved in sea-water and its adsorption on particulate material is given. Various methods of isolating dissolved organic matter, its effect on the buffer system and on calcium carbonate solubility are discussed. In addition some data on the fatty acid content of sea-water is given. The studies on the sorption of organic material on suspended sedimentary particles indicate a functional relationship of mineralogy and organic type.

Introduction

Fostered in part by the American Petroleum Institute an interest in the organic matter in marine sediments began to develop about 35 years ago. The concern of the original investigators and of the Institute revolved about the origin of petroleum. In recent years this area of investigation has received a new surge of interest. Although the origin of petroleum is still a very important and vital issue, the role of organic matter in the processes of sedimentation including the formation of calcium carbonate deposits, diagenesis of sediments, the accumulation of metallic ions in sediments and marine ecology is rapidly receiving increased interest.

Two of the questions involved in all these issues are: (1) how much and what type of organic material is dissolved in sea-water, and (2) how does this dissolved organic matter become incorporated into the bottom deposits.

The literature shows that there is approximately 1 to 5 mg of dissolved organic material per litre of sea-water. According to Plunkett and Rakestraw (1955) the distribution is not uniform. Much of the earlier work concerning the type of organic material in natural waters has been summarized by Vallentyne (1957). He points out that analytical methods such as chromatography and spectroscopy are now developed and being used so as to permit reliable investigations on the organic chemistry of sea-water. Various types of organic compounds have been reported in marine waters. Wangersky (1952) isolated what appeared to be a rhamnoside and also a dehydroascorbic acid. Hargreaves *et al.* (1951) demonstrated the presence of citric and malic acids in the sea. Vitamin B_{12} has been isolated from sea-water by Cowey (1956). Slowey *et al.* (1959) have characterized some organic material extracted from sea-water by ethyl acetate. These are some instances of reports on the types of organic matter to be found dissolved in sea-water.

This, however, is not merely a problem of analytical interest. It has now been demonstrated that the carbon dioxide in sea-water is affected by carbamino

* Contribution from the Department of Oceanography and Meteorology of the A. & M. College of Texas, Oceanography and Meteorology Series, No. 128.

carboxylic acid complexes (Smith, 1958). Also the solubility of calcium carbonate is affected by organic matter dissolved in the sea (Neuberg *et al.*, 1957; Park and Hood, 1959).

The organic materials found in the sedimentary deposits probably have a multiple origin. They may be deposited as discrete particles from a marine or terrestrial source. In addition they may be formed in place by the action of macro- and micro-organisms living in the deposits. Another mode of origin may well be the process of "scavenging" the dissolved organic matter from the water via sorption on the surface of settling inorganic particulate matter.

The work of the soil scientists and clay mineralogists has shown the great potential for mineral surfaces to adsorb organic materials (Evans and Russell, 1959; Lynch *et al.*, 1956, 1957; McLaren, 1954). Whitehouse (1955) showed that the uptake of carbohydrates varied with the mineral in suspension and the carbohydrate dissolved in the sea-water. He observed that montmorillonite removed more sucrose from solution than galactose or raffinose. According to his data kaolinite was the most efficient in removing galactose and sucrose from solution, but it failed to take up any raffinose. Illite was the least efficient in removing all three carbohydrates. Similar studies by Bader and Jeffrey (1958) indicated that for montmorillonite, kaolinite, illite and mixed marine sediments the former was the most efficient in removing undifferentiated organic compounds from solution. The source of the organic material in this instance was obtained by decomposing phytoplankton grown in a medium containing 0·025 μc of carbon-14 per litre in the form of $NaHCO_3$.

Basically the investigations discussed herein were initiated in order to determine the amount and type of organic matter in sea-water and also to gain an insight into one mechanism for removing this organic matter from the water and incorporating it into the bottom deposits.

The Organic Compounds of Sea-water

It is necessary that high purity organic concentrates be isolated from sea-water for quantitatively determining the soluble organic compounds present. However, since the ratio of organic to inorganic materials in sea-water is of the order of 10^{-4}, simple concentration is impractical. A study was thus undertaken to devise methods for separation of the organic constituents from the salt components of sea-water samples. To determine the effectiveness of these techniques, nutrients and carbon-14 labelled carbon dioxide components were added to natural cultures of sea-water. The cultures were allowed to grow for approximately 3 months, then placed in the dark for a period of 1 year. By this process labile organic materials decomposed and only those resistant to bacterial action remained in the water. Five methods for isolating the organic compounds were then evaluated (Jeffrey and Hood, 1960): (1) dialysis, (2) adsorption of the organic constituents on various materials, (3) ion exchange of the organic constituents on resins, (4) solvent extraction and (5) coprecipitation with inorganic salts. All methods were found to have both advantages and disadvantages (see Table 1). The results indicated that total organic material was best removed and isolated from sea-water by electrodialysis with a cellulose membrane, or by coprecipitation with ferric hydroxide.

To obtain data on the geochemical significance of the organic materials of sea-water, several 400-gal samples of surface sea-water were collected and coprecipitated by means of ferric and magnesium hydroxides. The precipitate was then dissolved in 3 N hydrochloric acid and the iron extracted by *iso*propyl ether. The iron was removed almost quantitatively, leaving behind materials not extractable in ether. The samples thus obtained had sufficient organic material for analysis.

Table 1. Summary of efficiency of various methods for isolation of organic material from sea-water

Method	Percentage total organic matter isolated	Advantages	Disadvantages
Electrodialysis with cellulose membranes	97	Ideal for very large molecules or colloidal micelles.	Inorganic sulphates not completely removed. Large volumes of water must be evaporated at a low temperature.
Carbon adsorption and elution with phenol	100 adsorbed 62 removed with phenol	All of organic constituents are absorbed.	The carbon must be refluxed with phenol to remove inorganic impurities from the carbon.
Coprecipitation with $FeCl_3$	100	All of organic constituents concd. by a factor of 10,000 in one simple operation.	$Fe(OH)_3$ must be separated from the organic fraction.
Solvent extraction A. phenol B. ethyl acetate C. 2·6-lutidine	 65 40 32	Quick and simple to prepare an organic concentrate without altering the organic material appreciably.	Any one solvent separates only a fraction of the organic matter.
Ion exchange	65	No particular advantage.	Too large an organic blank obtained. Impractically large volumes of resin are necessary to prepare a sample. Large amounts of liquid must be evaporated to obtain the organic concentrate.

Buffer effects

A concentrate of organic material representing 5 gal of sea-water shows substantial buffer effect when titrated with dilute sodium hydroxide solution. Such a titration curve is shown in Fig. 1. These data indicate that at least part of the organic compounds present are proton acceptors with the highest capacity existing in the pH range between 7·8 and 9·0. The biochemical compounds which

are most likely to contain replaceable hydrogen atoms are the carboxylic acids and the amino acid salts. The slight break in the titration curve which occurred at pH 3·5 may be due to carboxylic acids; however, the major break is believed to result from replacement of hydrogen on proteins, peptides or amino acids. Peptides are the more likely since relatively low concentrations of proteins and free amino acids have been found in these samples. The replaceable hydrogen may be functional in alkaline earth salt formation, a phenomenon known to occur with carboxylic acids and also carbamino carboxylic acid complexes (NEUBERG *et al.*,

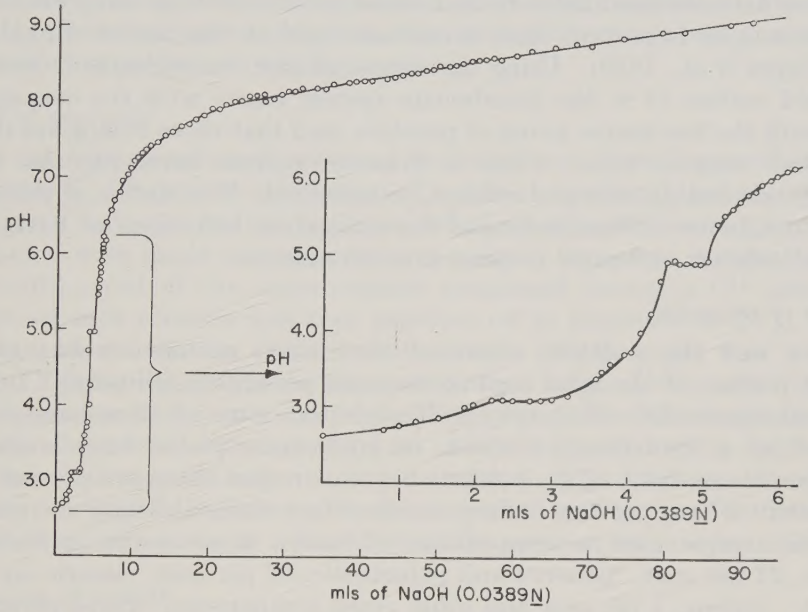

Fig. 1. Titration curve of organic material extracted from sea-water.

1957) and may also provide compounds necessary for carbon dioxide complex formation through the carbamate reaction. Further chemical analysis is needed to show the exact nature of these compounds.

$CaCO_3$ *complexes*

Because of the strong buffering effect of the organic material in the concentrates it was thought that these materials could be functional in calcium carbonate solubility relations. To investigate this point, calcium-45 in the ionic form was added to sea-water concentrate at a pH of 3·0. The pH was then increased to 8·5 by means of sodium hydroxide, and the resulting solution subjected to dialysis in a Viscose sausage skin. Comparison with a control containing only inorganic calcium-45 showed that the rate of removal of calcium from the solution containing organic material in concentrations likely to be present was much slower indicating the formation of complexes of calcium with the organic material in sea-water. An investigation of the use of ion exchange for chromatographic separation of the inorganic and organic forms of calcium is now under way.

Amino carboxylates in sea-water

In work with the form of carbon used in photosynthesis it was observed that certain marine algae (*Nitzchia closterium* and *Platymonas* sp.) when grown under autotrophic conditions in inorganic media showed preference for the amino acid carboxylates in sea-water to inorganic forms of carbon for photosynthesis (Hood *et al.*, 1959). These data indicate that dissolved organic material in sea-water is involved in the carbon dioxide equilibrium relationships and must be taken into account in any attempt to understand the kinetics of the carbon dioxide system. Quantitative data obtained indicate that concentration of these complexes may be several times more important than is carbonic acid to the carbon dioxide components (Smith *et al.*, 1959). Using the coprecipitated concentrates it was found that labelled carbon-14 in the bicarbonate system reacts with the concentrates, probably with the free amine group of peptides, and that these complexes do form soluble alkali mineral salts. Thus a dynamic system involving the organic constituents, carbon dioxide and calcium, is indicated. Elucidation of this system may lead to a better understanding of the anomalous behaviour of both carbon dioxide and calcium carbonate in natural environments.

Fatty acids of sea-water

Jeffrey and Hood (1960) observed that ethyl acetate would extract a substantial portion of the total organic material present in sea-water. Infra-red analysis and electrochromatography indicated that some of these materials are represented by a lipid fraction which, on hydrolysis, yields fatty acids. Gas chromatographic analysis of the extracts has shown that these are present to the extent of about 0·5 mg per litre in deep ocean surface water and may decrease with depth. One sample gave a composition of lauric, 6 per cent; myristic and myristoleic, 22 per cent; palmitic and palmitoleic, 52 per cent; stearic and oleic, 10 per cent; linoleic, 2 per cent and other trace components. These percentages are calculated on the basis that the acids present are those between eight and twenty carbon atoms in chain length.

Sorption of Organic Matter

The process of sorption of organic materials dissolved in sea-water by the suspended sedimentary particles may prove to be a very significant method of removal of such organic matter from the water and incorporating it into the bottom deposits. This process is far from simple, since factors such as temperature, salinity, pH, type and concentration of mineral particles and type and concentration of the organic matter are all variables well worthy of consideration. The research programme was developed with these in mind and with the realization that the availability of organic matter to decomposition, alteration and migration by diffusion or flow through sediments is related to whether it is free or bound to particles.

Kaolinite, montmorillonite and illite are three of the principal minerals under investigation and were selected for one or both of the following reasons: (1) because of the active surface of montmorillonite and illite and (2) since studies showed that the sediments emanating from the mouth of the Mississippi River

consisted of about 30 per cent of clay minerals in suspension. Montmorillonite comprises 50 per cent of the clays with kaolinite and illite 25 per cent each (McAllister *et al.*, 1958). Fine-grained quartz sand and calcium carbonate precipitates are also being used.

The organic compounds presently under investigation are carbon-14 labelled and include the following: (1) sucrose, (2) glucose, (3) fructose, (4) alanine, (5) acetic acid, (6) benzoic acid, (7) oxalic acid, (8) succinic acid, (9) stearic acid, (10) butyric acid, (11) aspartic acid, (12) glutamic acid, (13) valine and (14) glycine. All of these have been identified in natural waters and sediments (Vallentyne, 1957). As the investigation progresses the number of organic compounds, minerals and environmental factors will be increased.

Method of analysis

The minerals, montmorillonite and kaolinite, were first dispersed in 0·1 g/l. of Marasperse N. They were not cleaned of residual organic matter, thus the uptake values given here may well be minimal. Solutions of aspartic acid, glucose, sucrose and alanine were made at concentrations ranging from 0 to 3 mg/ml and spiked with 0·0025 μc/ml of the same organic compound having a C^{14} label. Half a millilitre of each solution was then pipetted off in duplicate at 20°C, dried in a planchet and counted in an automatic sample changing gas flow proportional counter. Fifty milligrams of the mineral was then weighed, wet with 1 ml of water, added to each solution and shaken for 20 min at 20°C. The suspended material was then allowed to settle out and $\frac{1}{2}$ ml of liquid pipetted off in duplicate. This liquid was then counted as previously mentioned. The difference in radioactivity between the original solution and the supernatant was used to determine the amount of organic material removed from the solution by the clay mineral.

Discussion of results

Fig. 2 gives ample evidence of the differential uptake of organic compounds by two minerals common to the bottom deposits of the Gulf of Mexico, namely montmorillonite and kaolinite. It can be seen that aspartic acid and montmorillonite have the greatest affinity in comparison to the other systems investigated. At maximum concentration (3 mg/ml) approximately 13 per cent of the aspartic acid was removed from the solution by montmorillonite. Kaolinite removed only about 2 per cent. It can also be observed that the aspartic acid–kaolinite combination attains the point of maximum sorption at much lower concentrations of dissolved aspartic acid. Relatively little sorption occurs beyond a solution concentration of 2·0 mg/ml. Even at the relatively high concentrations used (maximum 3 mg/ml) montmorillonite appears to have a continuing capacity for taking up the aspartic acid. It is also noteworthy that sequence in uptake is apparent for both minerals, namely aspartic acid, alanine, glucose, sucrose for montmorillonite and aspartic acid, alanine, glucose for kaolinite.

Though the data presented here are far from comprehensive there is an indication that the mineralogy of the particles settling through the water column will be in part a controlling factor as to the types of organic compounds supplied to the bottom deposits. It must be mentioned, however, that at present it is not

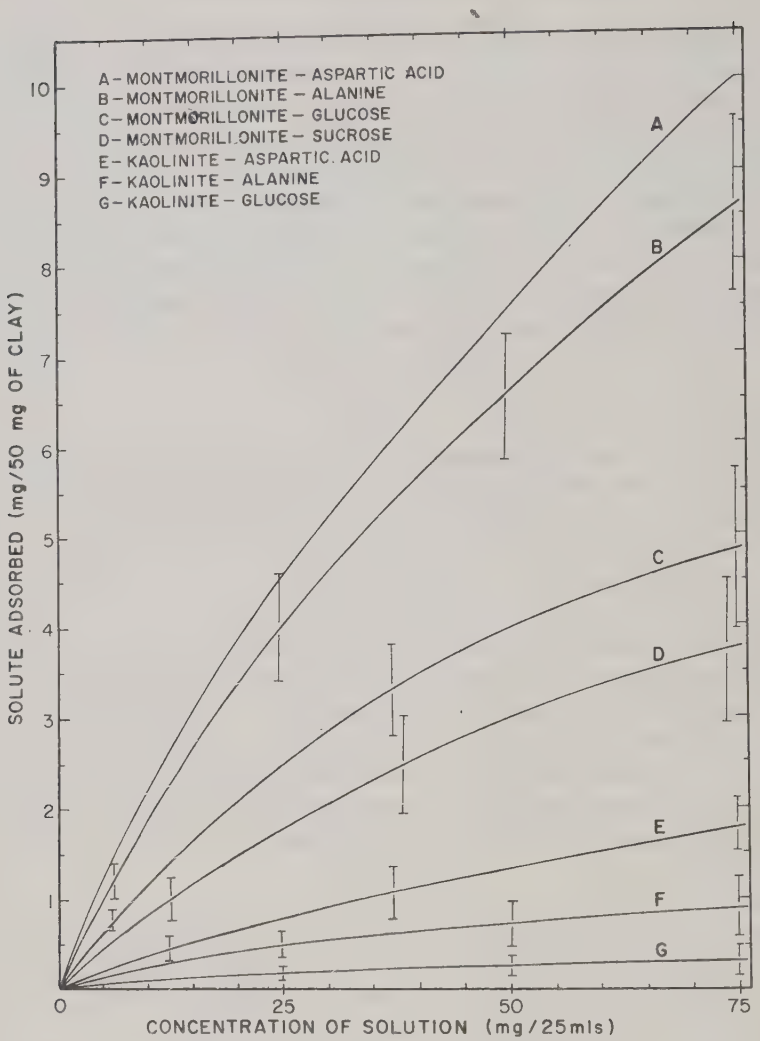

Fig. 2. Adsorption of organic compounds by clays.

certain whether this uptake is adsorption, absorption or a combination of both. In view of the fact that less than 2 per cent of the organic compounds associated with the clays can be removed by repeated washings it appears that chemical bonding is associated with the uptake phenomenon. Additional work is necessary in order to elucidate this. Also, temperature and salinity effects must be known. This work is now in progress.

Acknowledgements—The investigations on dissolved organic matter in sea-water and associated problems were supported by the Robert A. Welch Foundation, grant no. 022 and National Science Foundation grant no. 5038. The study on sorption of organic matter was supported by the U.S. Atomic Energy Commission, contract no. At-(40-1)-2061. Appreciation is extended to Dr. W. A. White, Illinois State Geological Survey; National Kaolin Products Company; and American Colloid Company for supplying the deposit clays for this study.

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Geochimica et Cosmochimica Acta, 1960, Vol. 19, pp. 244 to 260. Pergamon Press Ltd. Printed in Northern Ireland

Bacterial activity in sediments of shallow marine bays

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Abstract—Complete mineralization of organic matter results from a multitude of complex bacterial activities which must involve more than one species of bacteria. The diverse bacterial activities influence the pH, redox potential, production of gases and their resulting disfiguration of the sediment, concentration of certain elements, precipitation and solution of carbonates, diagenesis of silicates, the production of complex organic compounds such as surface-active agents and many other properties of sediments. The bacterial activities are not constant but proportional to the amount of organic matter, pH, sediment particle size, temperature, sequence of physical changes caused by the bacteria in the sediment and the sequence of the production of organic by-products during decomposition.

Introduction

When one takes a short field trip over the wide expanse of mud flats adjacent to the shallow marine bays near the Institute of Marine Science at Port Aransas, Texas, it is apparent that the sedimentary environment is not uniform and that almost daily mechanical, biological and chemical changes are taking place. These continual changes must ultimately influence the geochemistry of the environment. When the sediment is finally buried, compaction and other diagenetic processes will continue. The important point here is that bacterial activity in freshly accumulated sediments will have a profound direct or indirect effect on many subsequent sedimentary processes. In a review of the significance of marine microbiology, Wood (1958) includes a paragraph on geological considerations and makes the statement "The relationship between micro-organisms and geological processes has been the subject of some wishful thinking, in that geologists have tended to assign to biological agents many things which they could not readily explain, and microbiologists have not been as helpful as they might have been in suggesting how micro-organisms influence the environment. The latter question is important as the organisms are ephemeral and can only be known from the past by their effect on the milieu in which they once lived."

The following thesis is an attempt to point out that the bacteria in the surface 6 in. of the sediments of shallow marine bays near Corpus Christi, Texas (Fig. 1), may be primarily responsible for much of the diagenesis taking place. The great change in the consistency of the bay sediments, within a few feet of surface area or a few inches of depth, indicates that microbial activity will vary considerably. The writer makes no claims of completeness of all the bacterial activities but will emphasize some aspects which are important to sediment diagenesis. Various activities of bacteria are discussed separately for the purposes of clarification and the reader must therefore overlook the fact that repetition may occur.

The Sedimentary Environment

To understand better the changes which take place in sediments the sedimentary environment must be defined. The term sedimentary environment was adopted in this paper to relate different characteristics of sediments to biological

activity and includes combinations of : location within the bay, aerobic conditions, anaerobic conditions, different amounts and types of organic matter, mineral content, grain size, skewness or grading of clastic particles, pH, etc. Each environment may be capable of supporting a given population of living organisms and in turn, the organisms will be capable of changing the environment during their activities. If the area is smaller than a few square centimetres the term microenvironment is used.

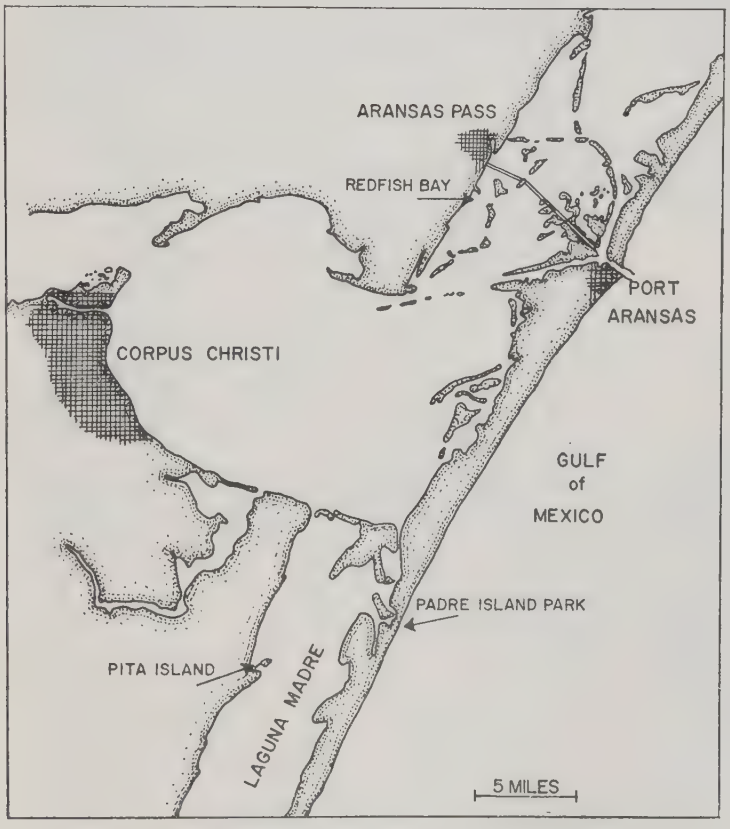
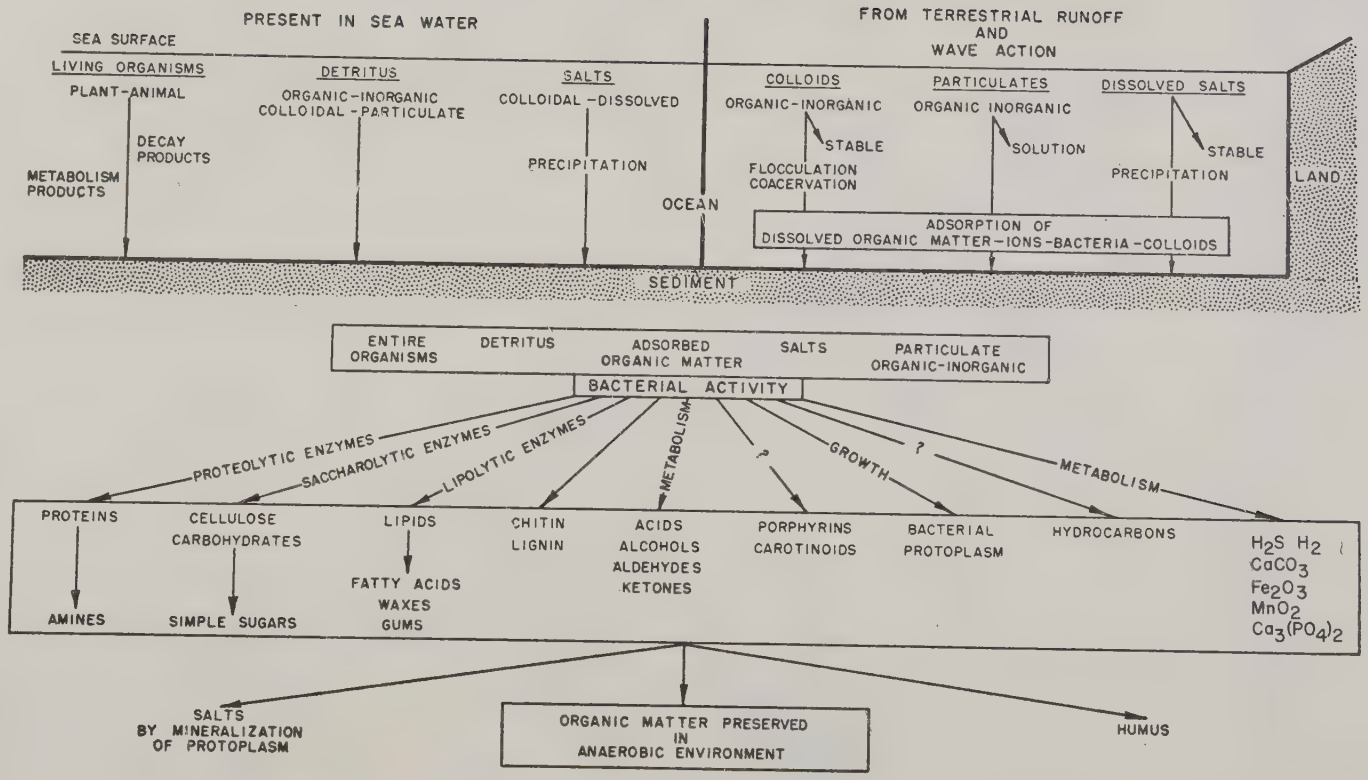


Fig. 1. Shallow marine bays near Corpus Christi, Texas.

The sedimentary environments discussed here are restricted to typical bays which extend inland several miles, have extensive shore lines and reef structures, are separated from the Gulf of Mexico by barrier islands, have water depths of less than 15 ft, salinities from near fresh-water to hypersaline and little communication with the Gulf of Mexico. The water is alternately stirred by the violent "northers" in the winter and the constant strong southeasterly trade winds during the summer. Most of the tide is caused by wind action, as the average lunar tide of $1\frac{1}{2}$ ft extends only a short distance into the bays from the occasional passes through the barrier islands. Along the edges of the bays there are extensive mud flats, as shown in Fig. 2, which are alternately covered with water or are dry during changes in wind direction. Shepard and Moore (1955) described the area.



BACTERIAL ACTIVITIES IN SEDIMENTS

Fig. 3. Bacterial activities in marine sediments and processes of sedimentation.

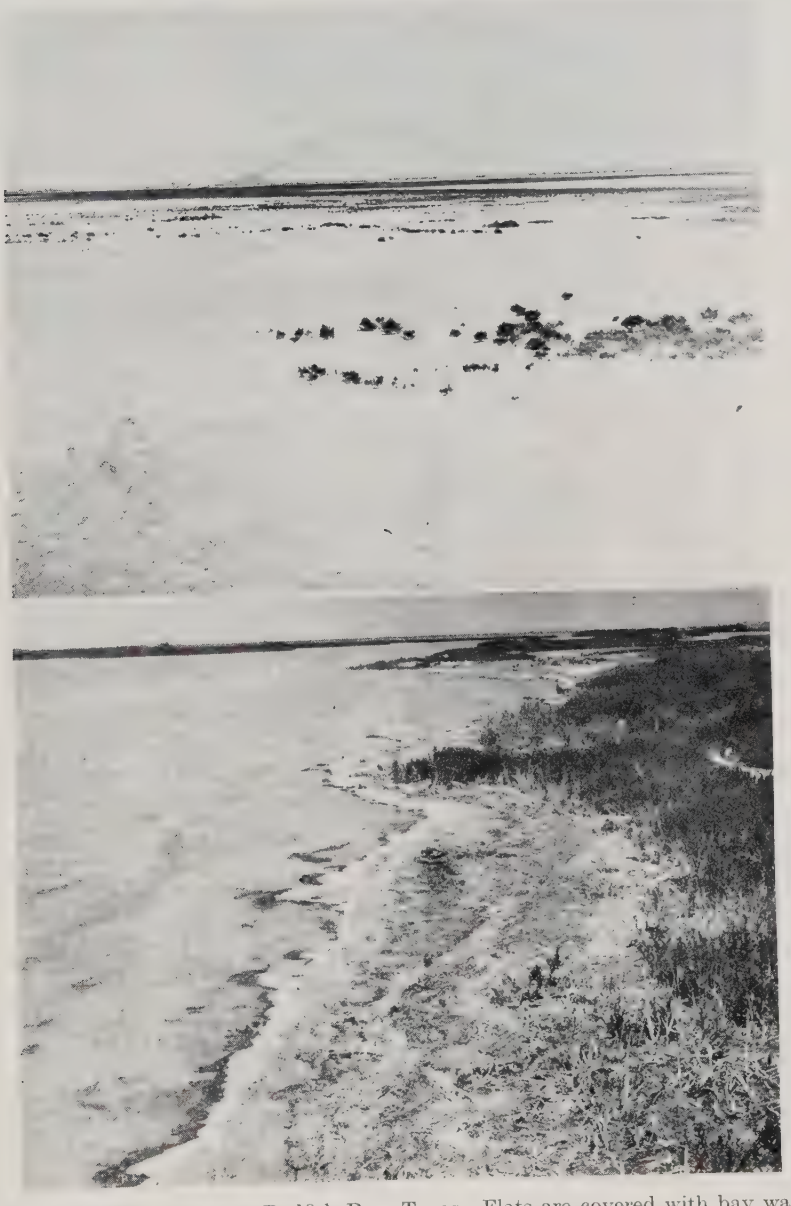


Fig. 2. Mud flats adjacent to Redfish Bay, Texas. Flats are covered with bay water at irregular intervals. The flats may or may not be covered with vegetation. The wind blows organic material on the shores in irregular patterns.

The physical action of the winds and tides in the shallow bays constantly acts on the surface of the sediment. The wave action in the bays, produced by strong winds, often moves enough sediment to change its level 2 to 4 cm. After a storm the suspended sediment settles to the bottom, or on benthic plants from which it is slowly washed off to the bottom. In certain areas of the bays the settling rate of suspended matter is very rapid, within 24 hr, while in other parts of the bays the particles remain in suspension for days or weeks. This clearing phenomenon has not been explained, although certain speculations have been made that the plants stimulate flocculation or that an abnormally high shell-fish population acts as a biological filter.

Fine materials and organic matter are deposited in the lee side of bays and in shallow water rendered calm by the presence of aquatic plants. Sand and shell are common in areas where water motion washes out the fine material. The edges of bays may have discrete patches of shell, sand, silt or clay within a few feet of area or a few inches of depth. The centre of the bays are somewhat more uniform but also vary considerably with oyster reefs, grass flats and sand bars.

The organic matter in any area of the shallow bays cannot be directly attributed to any one source. The distribution of types of organic matter in sediments will be related to the types of living organisms and particulate organic matter which is present in the water above or attached to the bottom (Fig. 3). Larger pieces of organic matter may be directly added to the sediment, creating abnormally high localized concentrations. It is interesting to note that living organisms contain an average of from 5 to 10 per cent organic matter and thus sediment consisting of large masses of living organisms will not have abnormally-high concentrations of organic matter. Shepard and Moore (1955) found an average of 4 per cent organic matter in the shallow bays near Rockport, Texas.

The processes of sedimentation are also quite important in the diagenesis of the sediments (Fig. 3). Small particles scavenge organic matter, other inorganic ions, and micro-organisms by surface attraction before settling to the bottom (Fox, 1955). Inorganic compounds are added to sediment, adsorbed to particles by chemical precipitation, by iron or magnesium hydroxide scavenging or in the remains of living organisms (Goldberg, 1954, 1957). Sediments will be differentially graded by the physical action of waves and thus the intensity of the wave action will produce layering of different particle sizes containing organic matter and bacteria. Terrestrial rivers and runoff from watersheds constantly bring new organic and inorganic materials into the bays. These materials are mixed with sea-water and chemical precipitation and flocculation occurs. Seasonal layers of material are deposited.

Collier and Hedgpeth (1950) state that the temperatures of the shallow bays and sediments follow the air temperatures closely. During the winter the temperatures may be as low as 4°C and during the summer as high as 30°C. However, some shallow water areas and exposed sediments covered with blue green algal mats may reach much higher temperatures than stated by Collier and Hedgpeth. In Table 1 are listed temperature profiles in sediments covered with blue green algal mats and adjacent uncovered sand.

The warm temperature of the surface sediments exerts a great influence on the

activities of the living micro-organisms. van't Hoff's law states that each 10°C increase up to 40 or 50°C increases the rate of specific biological activities two- or threefold. Wheatland (1954) found that the rate of formation of sulphide increases with temperature, doubling approximately for each 10°C rise.

Table 1. Temperatures in sediments of Redfish Bay at 11 a.m. April, 1959

Sediment depth (cm)	Temperature (°C)		
	Area 1*	Area 2†	Area 3‡
1	34·5	—	33·0
2	34·5	30·0	33·0
4	34·0	30·0	33·5
6	32·5	28·5	32·5
8	31·0	27·5	31·0
16	26·5	24·5	26·5
Air	32·0	32·0	32·0

* Area 1 was selected in sediment approximately 20 m from the water's edge in a blue green algal mat.

† Area 2 with no blue green mat was approximately 3 m away from area 1.

‡ Area 3 in soft sand was approximately 20 m away from area 2.

Bacterial Populations

It is impossible at present to determine the total numbers of individual bacteria in sediments by direct microscopic observations because the small non-motile bacteria cannot be readily distinguished from the finer particles of detritus or clay minerals. Usually specific types of bacteria are determined by the plate count procedure. However, a different type of media is required for each different type of bacteria, and it is impractical to devise media which will provide for the detection of all the bacteria. Surface sediments may have up to 100 million bacteria per g, which can be enumerated on nutrient sea-water medium (Volkmann, 1959, personal communication). If one assumes that the weight of an average bacteria $1 \times 2\ \mu$ in size is approximately 10^{-11} g, 100 million bacteria will weigh 10^{-3} g or represent 1·0 mg/g of sediment. Minimum bacterial populations by dilution counts on several media have been reported by Oppenheimer (1952) for Aransas Bay, Texas a few miles north of the Institute. Table 2 gives the numbers of bacteria at different sediment depths.

Migration of Bacteria

Bacteria are easily transported through the water and sediments by currents either suspended or adsorbed to larger floating or particulate materials. When storm waves remove the surface sediments from the bottom of shallow bays, the indigenous bacteria are washed into the water column and the sediment particles may adsorb new bacteria from the water before settling back to the bottom.

Many marine bacteria are mobile and can move through sediments, independent of density currents. Oppenheimer (1952) determined the migration of two species

of marine bacteria through sterile marine mud untreated and enriched with nutrients (Table 3). The results indicate that although bacteria can move through sediments, the migration rate is influenced by the presence of nutrients as also reported by DEBYSER (1951). ZOBELL (1947) reported that sulphate-reducing bacteria migrate at average rates exceeding 1 in. per day in nutrient-containing sand.

Table 2. Minimum bacterial populations by dilution culture counts on sediment samples from Aransas Bay, Texas, January, 1952

Core no.	Depth of sediment (cm)	Number of bacteria per ml of sediment			
		Aerobes	Starch splitters	Anaerobes	Sulphate reducers
A-46-B	1	500,000	500,0000	10,000	1000
	40	1000	10	50	50
	100	50,000	500	500	100
A-47-B	1	1,000,000	500,000	10,000	5000
	40	50,000	10,000	500	0
	130	50,000	10,000	500	0
A-49-B	1	5,000,000	500,000	50,000	1000
A-56-B	1	5,000,000	5,000,000	5000	1000
	40	10,000	5,000,000	1000	0
	75	5000	500	1000	0

Table 3. Migration of bacteria in marine mud

Organism	Migration rate (cm/day)	
	Untreated	Nutrients added
Pure culture:		
Serratia marinorubra	1·2	3·9
Desulfovibrio sp.	2·2	1·9
Mixture of two:		
Serratia marinorubra	1·1	3·4
Desulfovibrio sp.	2·7	2·9

BACTERIAL ACTIVITIES

Some bacterial activities in sedimentary processes are illustrated in Fig. 3. The large number of bacteria in the sediments immediately start to decompose the organic matter into various intermediate organic compounds, or carbon dioxide and inorganic compounds (WAKSMAN *et al.*, 1933).

Many of the bacteria in sediments are heterotrophic and require organic matter for energy and growth. Therefore, the amount and type of organic matter in

sediments will directly affect bacterial activity and, in general, the greatest bacterial activity will occur in areas where the greatest amount of available organic matter is present.

The most important function of bacterial activity is the mineralization of organic matter produced by plants and animals. Fig. 4 illustrates the role of bacteria in the cycle of organic matter in a marine environment. The inorganic materials assimilated by plants and stored by plants and animals are released,

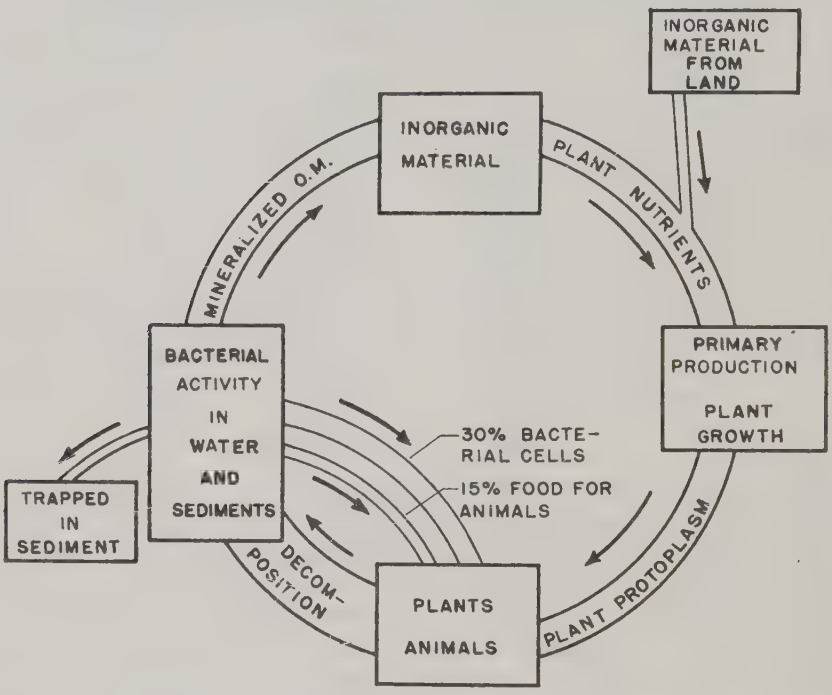


Fig. 4. A simplified cycle of organic and inorganic matter in a shallow marine bay.

after death, by bacterial decomposition. The end products and pathways of decomposition are controlled by the types and number of bacteria present. Decomposition is more complete when more diverse bacterial types are present. Total numbers of bacteria alone may not indicate the possible degree of mineralization. Bacteria may directly convert into their own cell protoplasm from 20 to 40 per cent of the organic matter they utilize. Part of the organic matter such as lignin, chitin and pyrroles (Barghoorn, 1952) may be in a form difficult or impossible for bacteria to decompose. Some organic decomposition products (estimated at 15 per cent in our bays) may be directly consumed by other plants and animals which live in the sediments.

Generally, it has been assumed that bacteria will decompose fats, proteins and carbohydrates before other complex organic compounds (Hulcher *et al.*, 1957). However, Abelson (1956) has found amino acids present in fossil shells and Collier *et al.* (1953) found relatively large concentrations of free sugars in marine

sediments. These reports indicate that either amino acids or sugars are intermediates of current metabolic processes, which appears unlikely, or that the compounds may persist for longer periods of time than has been anticipated.

Rate of Decomposition

Decomposition of organic matter in sediments does not proceed at a uniform rate and appears to be somewhat related to sediment particle size and season. The rate of decomposition is faster during warm months but continues during the cold months. Sand sediments stored at 4°C for 40 days may lose as much as 95 per cent of the total organic matter (Volkmann and Oppenheimer, 1959) while clays may lose only up to 75 per cent. One would expect decomposition to be more rapid in sand than in clays because of the greater abundance of types of bacteria in the former. During the winter, in the sand along the shores of the bays, the organic matter is lowered to 1 per cent or less by bacterial oxidation. The difference in decomposition rate in sand and clay may account for the observations that ancient clays contain more organic matter than sands. Volkmann and Oppenheimer (1959) find that certain surface sands contain more organic matter than clays. They attribute this to the large number of photosynthetic and other micro-organisms inhabiting the upper 1 cm of sands where light can penetrate.

Bacterial Activity and Particle Size

A direct correlation between bacterial activities and depth of sediment is possible only when sediments are uniform. Bacterial activities may be directly influenced by the particle size and the interstitial space of sediments. Direct microscopic observations show that the number of types of bacteria decrease, and ZoBell (1946a, p. 94) shows that total numbers of bacteria increase with a decrease in particle size. Recently the writer has measured the bacterial population in sediments and found variations between 10,000 and 100,000,000 cells per ml within a few centimetres of depth or lateral area. It is well known that bacteria grow better where a large surface is available (ZoBell, 1946a, p. 84). Also sediments adsorb organic materials by surface attraction, and smaller particles with larger potential surface area will adsorb more material than coarse sands, thus providing more or less nutrients for bacterial activity. Bacteria and their enzymes are also attracted to the surface of particles, the net result being that the bacteria and organic matter are concentrated together. The same phenomenon will remove toxic materials from bacteria suspended in water.

The large interstitial spaces of sand or shell provide a living space for most types of bacteria and allow free movement of the organisms by motility and density currents. Clays with particles of 1 μ or less have very small interstitial spaces (unless highly hydrated). The small pore spaces afford living room for only the smaller types of bacteria and the migration of the bacteria and the diffusion of food and metabolic wastes are inhibited (Oppenheimer, 1960).

In Fig. 5 are two photomicrographs taken of representative sand and clay sediments from the Laguna Madre, Texas area. Such photographs are difficult to obtain because of the optical limitations in depth of field. However, the photographs

clearly indicate that the interstitial spaces of the sand contain many types of living micro-organisms. Most of the smaller particles have been identified as bacteria. The picture of the clay represents a magnification of X2000 and most of the particles are less than 1 μ in length. It is difficult to recognize the bacteria, and those that can be recognized are less than 1 μ in size and are only short rods and cocci.

Clay Adsorption

Kuenen (1950, p. 400) states that organic matter increases with a decrease in grain size and attributes this to the protective action of clays and the porosity of sand. Carrigy (1956) indicates that topography is important in the distribution of organic matter and smaller particle size sediments contain higher organic matter The results shown in Fig. 5 indicate that the small interstitial spaces of the clay may be more important because of the limitation of types of bacteria.

However, one must not fail to distinguish between available organic matter and that adsorbed in the clay lattice where it might be unavailable to the bacteria or enzymes. Lynch and Cotnoir (1956) report that bentonitic clay attenuated the breakdown of many organic compounds while illite and kaolinite had no effect. Mortland and Gieseking (1952) show that inhibition of enzymatic phosphatase activity is roughly proportional to the base exchange capacity of the clays. Schwartz (1959, personal communication) has found that bacteria are selectively adsorbed on clay minerals and may be retained or replaced by other bacteria in accordance to the net surface charge on the bacteria and surface attraction by the clay minerals.

Organic Matter Trapped in Sediments

Part of the organic matter produced in shallow bays (Fig. 4) is permanently trapped in the sediment and may be considered a source of fossil organic matter. Smith (1954) has shown that shallow water marine sediments in Laguna Madre contained approximately 400 p.p.m. hydrocarbons. An interesting calculation can be made from the data of Odum and Hoskins (1958), who have measured the primary productivity of the shallow bays and found an average total production of 5 g/m^2 per day or 1825 g per year, and Volkmann and Oppenheimer (1959) who report that some bay sediments in the same area contain an average of 5 per cent organic matter. If it is assumed that a deposition rate of 19 cm./100 years exists (Shepard and Moore, 1955, p. 1571), the calculated loss of organic matter to the sediment is 96 g year^{-1} m^{-2} or 5·2 per cent of the total organic matter produced. For life to continue in the bays, the loss of organic matter to the sediments must be replaced by equivalent nutrients from rivers, rain run-off, etc.

Trapped organic matter may not be released from the sediment for long periods of time, perhaps years, because of climatic conditions and the amount of mineralized materials available to the water will fluctuate and be reflected by all living organisms in the environment. The stirring effect of the waves on the sediment during storms will resuspend some trapped organic matter and release inorganic materials from decomposed organic matter to the water. Density currents and capillary action within the sediments may carry decomposition products to the mud surface. A part of the organic matter eaten by animals is

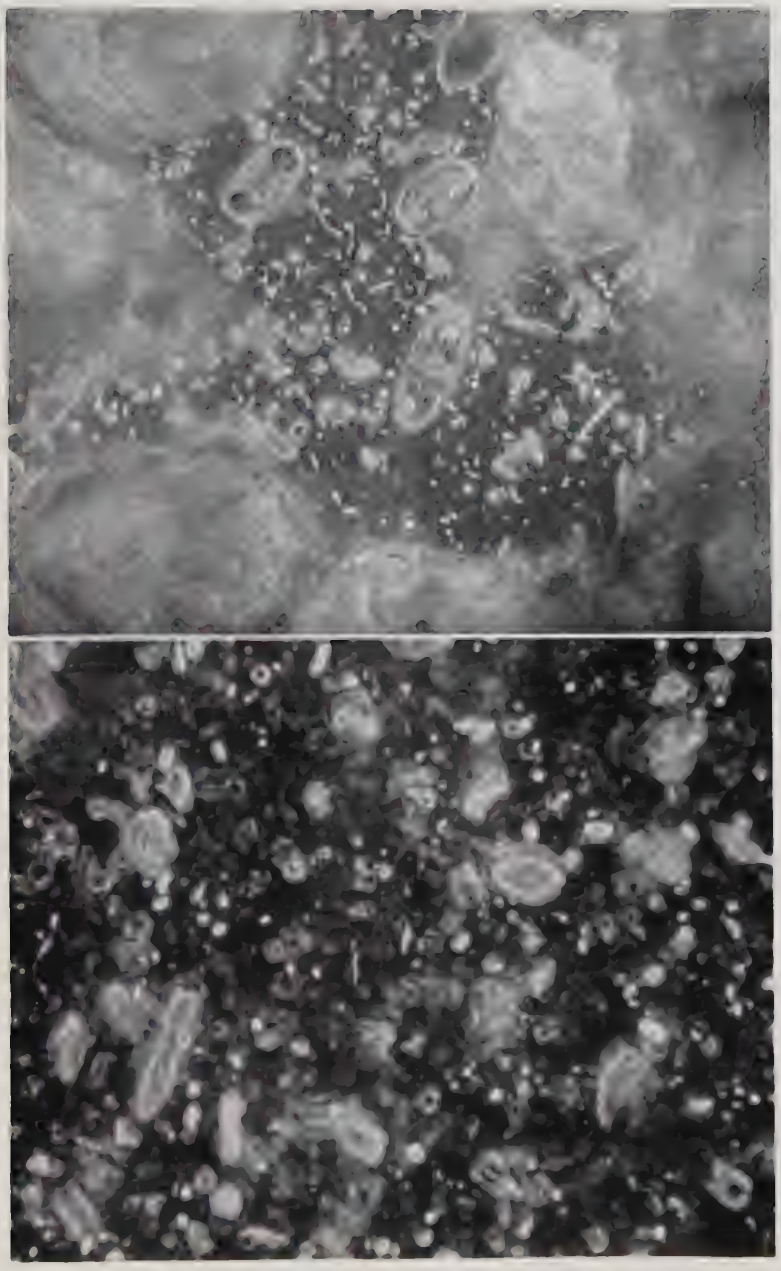


Fig. 5. Phase photomicrographs of surface sediments from the Laguna Madre, Texas tidal flats. (A) Sand. ×250. (B) Consolidated clay. ×2000.

consolidated into faecal pellets which, in some sediments, appear to be somewhat resistant to decomposition.

pH and Redox Potential

As one might expect, the pH and Eh of the sediments of the shallow marine bays do not conform to any trend or general rule. In most sediments, bacteria and other living organisms are the sole agents responsible for anaerobic conditions and changes in pH. Because of the diversity of sediments and types of bacteria, the pH and Eh at any position in the sediments are a function of the net balance between the types of bacteria, organic matter, the buffering and poising capacity of the sediments and the diffusion rate of O_2 through the sediment. Certain organic compounds are reducing agents, but the state of reduction is a function of bacterial activity. Thus, a simple organic compound such as pyruvic acid acts as both oxidizing agent or reducing agent depending on the biological forces, namely enzyme systems, which use pyruvic acid as a substrate. When a plant or animal dies, bacteria start to decompose it. The end products of decomposition will be reflected by the types and numbers of bacteria. Usually, O_2 is consumed during the decomposition of the living organism, and if the source of O_2 is limited, the environment becomes reduced. In this reduced state, organic matter is further decomposed by anaerobic oxidation involving electron transfer rather than molecular oxygen.

ZoBell (1946b) gives an excellent treatise on the redox potential of marine sediments in which he outlines the methods used for such studies and illustrates the complex chemical systems which are activated by changes in Eh. During a study of fresh-water lakes, Mortimer (1941) showed how the Eh in sediments had an annual vertical fluctuation which was apparently caused by physicochemical and biological changes during the season. Such changes are also evident in the shallow marine bays where the depth of the anaerobic zone is deeper in the sediment during the summer than in the winter.

Baas Becking and Wood (1955) have published Fig. 6 which illustrates how pH and Eh influence living populations of micro-organisms. The diagenesis of the sediments is influenced both by the changes in pH and Eh which micro-organisms produce in the environment and also by the metabolic activities of new populations which are established in the environment.

The variability of the pH and Eh in the shallow bays may be pointed out by Table 4 (taken from Oppenheimer and Kornicker, 1958) which shows profiles of pH and Eh for several locations in the bay sediments. Fig. 7, from the same paper, illustrates how pH will increase when the anaerobic sediments are removed from their *in situ* position. The low pH is thought to be the result of the bacterial production of CO_2 and H_2S during decomposition of organic matter.

It is evident that many chemical changes take place in a localized area of sediment during the decomposition of a large piece of organic matter, such as an entrapped fish. First O_2 is consumed, then sulphide is produced by anaerobic bacteria and the pH may decrease. As decomposition proceeds, the readily available organic matter is used up and soon the bacterial activities begin to decline. When the O_2 demand is less than the rate of diffusion into the area,

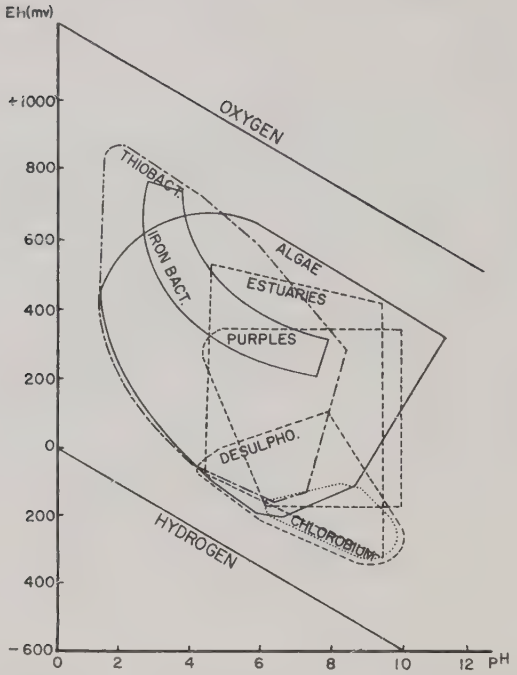


Fig. 6. Effect of pH and Eh on living populations of micro-organisms. (Taken from Baas Becking and Wood, 1955).

Table 4. *In situ* values of pH and Eh (in mV) in marine sediments near Corpus Christi, Texas and Sapelo Island, Georgia (taken from Oppenheimer and Kornicker, 1958)

Location		Depth of measurement (cm)					
		Surface	1	2	4	6	8
Aransas Bay near	pH	9·0	7·9	6·6	6·6	6·6	6·6
Corpus Christi	Eh	—	+144	+44	+44	—	−276
Aransas Bay near	pH	9·2	8·6	7·6	6·8	6·7	6·7
Corpus Christi	Eh	—	+4	−156	—	—	−300
Aransas Bay near	pH	8·6	—	—	6·6	—	6·5
Corpus Christi	Eh	—	—	—	−136	—	−256
Aransas Bay near	pH	8·4	—	8·0	6·9	6·5	6·5
Corpus Christi	Eh	—	+74	−36	−36	−146	−236
Corpus Christi	pH	8·0	7·1	—	6·8	6·7	6·7
Bay	Eh	—	−186	−286	—	—	−306
Mustang Island	pH	8·4	7·1	7·0	6·7	6·8	6·7
	Eh	—	−226	−256	−156	−56	−91
Sapelo Island	pH	8·2	6·9	—	6·3	—	5·9
Station 1	pH	8·4	—	6·8	5·4	—	5·4

Eh was not measured at Sapelo Island. Abundant H_2S was present and it was assumed that the sediment below the surface was anaerobic.

aerobic conditions will return and the pH will rise. Siebert and Schwartz (1956) have published information on the changes in sea-water containing *Fucus* and *Laminaria* during a 12 month period, which indicated that the Eh steadily decreased while the pH increased from 7·2 to 8·4.

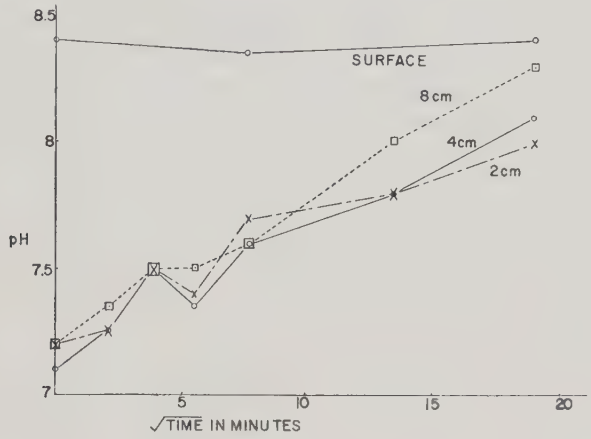


Fig. 7. Change of pH with the square root of time for a core removed from anaerobic sediment on Mustang Island, Texas. Measurements were made at the indicated depth from the top of the core. (Taken from Oppenheimer and Kornicker, 1958).

Sulphur Diagenesis

Sulphur plays a predominant role in the geochemical activities of micro-organisms in our marine environments. Sulphate is one of the most abundant ions in sea-water or approximately 7 per cent of the total salts. In anaerobic sediments, with energy derived from organic matter, the sulphate-reducing bacteria reduce sulphates to sulphides. In the presence of O_2, sulphur bacteria oxidize the reduced sulphide to sulphur or sulphate (ZoBell, 1946b). During the change from the oxidized to the reduced state, considerable diagenesis of minerals may take place. Smith (1954, p. 392) states that algal muck from Laguna Madre with large amounts of H_2S contained 2·13 wt. per cent organic carbon and 7·54 wt. per cent S. Obviously S is being concentrated and probably is the result of bacterial decomposition of the organic matter. In adjacent aerobic sediments 3·74 per cent organic matter and only 1·94 per cent S was found.

Jones and Starkey (1956) have shown that S from sedimentary environments is depleted in S^{34} by the activities of sulphate-reducing bacteria. No fractionation of the S isotopes was noted during the oxidation of elemental S to sulphate by *Thiobacillus*.

Concentration and Migration of Trace Minerals

The change in pH caused by bacterial activities in sediments will affect many chemical reactions. Carbonates and phosphates may be dissolved or precipitated (LaLou, 1957; Macpherson *et al.*, 1958). The authors found that silica diatom shells in anaerobic sediments from Redfish Bay disappeared when the sediment became oxidized and the pH increased to 9.

Most living organisms concentrate some trace minerals during their growth (Vinogradov, 1953; Goldberg, 1957). When the remains of living organisms enter the sedimentary environment, decomposition takes place and concentrated minerals are released or are again concentrated in the cells of the bacteria. Arrhenius *et al.* (1957) found that fish debris from pelagic sediments contained a high concentration of cations whereas living fish from the surrounding area did not. Thus, the cations were concentrated after the death of the fish. The concentration may have taken place by a scavenging effect of the H_2S produced during the decay of the fish. Fishes decomposing in sediments are soon surrounded by an anaerobic environment even if they are lying on the sediment surface. Hydrogen sulphide is abundant as evidenced by the black colour of FeS. Arrhenius *et al.* (1957) states that the cation concentration process takes place in an early phase of sedimentation. This would certainly agree with the hypotheses that the cations were concentrated by the presence of H_2S during the anaerobic decomposition of the fishes.

The reducing environment produced by bacteria may also have an effect on the transportation of cations by turbidity currents and compaction through sediments. Many compounds such as hydrated Fe_2O_3 and MnO_2 or their carbonates, which are insoluble in the aerobic state, will be changed to soluble compounds in anaerobic conditions where sulphide is not present. Cations scavenged by the oxidized hydroxides or carbonates will then be released to the sediment and can be transported by currents or diffusion. When the soluble iron or manganese moves to an area where O_2 is present, precipitation occurs with a resulting concentration effect. If the redox-interface is in the water, the iron or manganese will form colloidal hydrated oxides which may again act as scavenging agents.

The dissolved iron and manganese may migrate through anaerobic sediments and be concentrated in certain areas where sulphide is present. In the presence of a localized excess of CO_2 from decomposing organic matter, MnS may be converted to the bicarbonate which is soluble and can diffuse away from the iron (Rankama and Sahama, 1950, p. 651). The soluble Mn^{++} may be oxidized by the activity of bacteria or by the presence of O_2 in aerobic conditions.

Kalinenko (1946) has reported that *Bacterium precipitatum* is responsible for the precipitation of ferro-manganese concretions of microscopic size in the Karskoje Sea.

Sperber (1958) has shown that the presence of H_2S will liberate phosphate from $Fe_3(PO_4)_2$ by the precipitation of iron as FeS. The soluble phosphate ions were then adsorbed by various adsorbants. It is very possible that the soluble phosphate in shallow bays may have been released in anaerobic conditions and adsorbed to sediment particles which are transported into the water during storm action. This adsorbed phosphate may be available to the plants. Harvey (1955, p. 43) mentions that strong winds which stir the bottom deposits will release a significant quantity of phosphate into the water above.

The method of conversion of FeS to FeS_2 (pyrite or marcasite) in surface sediments is not known. Emery and Rittenberg (1952) found that pyrite increased in proportion to hydrotrollite (FeS) and melinkovite (FeS_2) with depth of sediment. They suggest that the absence of pyrite in recent sediments indicates that the

pyrite has not had time to form from FeS. Emery and Rittenberg also state that other workers believe that pyrite is formed in an acid sediment and that marcasite is formed in an alkaline environment. Debyser (1955) found pyrite in recent sediments where H_2S was abundant and the pH was below 7. The writer has failed in laboratory experiments to produce pyrite (FeS_2) from sulphite derived from the bacterial reduction of sulphate.

Large concentrations of pyrite nodules are found in Cretaceous chalk near Austin, Texas and other areas. It is believed that pyrite has replaced the remains of organisms which were trapped in the sediment. One can estimate how much FeS can be produced from organic matter. Miller (1949) published data which shows that sulphate-reducing bacteria are able to produce 2·5 g of H_2S from 21 g of sodium lactate. If the H_2S combines with iron, 4·2 g of FeS would be produced or a ratio of 5 : 1 for lactate/FeS and 8 : 1 for lactate to H_2S. Senez (1951), using Warburg techniques, found that bacterial conversion of sulphate to sulphide required 2 moles of lactate for 1 mole of H_2S which gives a weight ratio of 5 : 1. A fish of 1000 g will have approximately 200 g of organic matter; fish are usually about 80 per cent water. If a conversion factor of 5 : 1 is used (which is undoubtedly too high for environmental conditions), the bacterial activity can produce about 40 g of FeS from the 1000 g of fish. This would be a considerable reduction in volume from that of the original fish, especially if pyrite is present which would require a second atom of sulphur for each iron atom. Thus, a pyrite mould or concretion of an organism buried in sediment would require additional iron and sulphur derived from another source than from energy from the fish organic matter.

Bacterial Colour Banding of Sediments

Along the shores of the bays, sediments are often deposited in layers. The distribution of organic matter in the sedimentary layers will not be uniform and thus, bacterial activities will not be uniform, a point already discussed in the decomposition of organic matter. The differential activities of bacteria within the sediment layer can produce alternate oxidizing and reducing areas. Sulphate-reducing bacteria in the anaerobic areas actively reduce sulphate to sulphide employing energy derived from the decomposition of organic matter. Fig. 8 illustrates the typical sand sediment structure found along one edge of Redfish Bay. The sediment photo was taken of an area approximately 30 m from the water's edge. Alternately the sample area is dry or covered by high tide. The sediment is banded by layers of sand and clay. The clay, which appears a faint grey colour, is almost undetectable within the oxidized layers from the surface to 8 cm depth. Below 8 cm the banded layers appear either black due to FeS or orange due to the presence of concentrated $Fe(OH)_3$ where the presence of oxygen oxidizes the ferrous sulphide to insoluble hydroxide. The presence of sulphide effectively concentrates soluble iron. The difference in the porosity or permeability of the sedimentary bands, and possibly turbidity currents, allows the lateral migration of oxygen into the different layers as shown at the profile depth of 20 cm where a shell-sand layer exists. When the sediment profile was exposed to the air, the black colour faded and was replaced by grey and orange layers as the FeS layers were oxidized to $Fe(OH)_3$.

Surface-Active Materials

One of the least studied of the bacterial activities in sediments is the production and destruction of surface-active agents. One can speculate here and mention that such organic materials are responsible for the hydration of sediments as well as the transport of particles in foam and surface slicks. Bacteria are able to produce and destroy surface-active agents, and in so doing affect compaction of hydrated and thixotropic sediments. La Riviera (1955) has reported that *Desulfovibrio* produces materials in synthetic media which decrease surface tension and which may affect the migration of oil through sediments. Von Riesen (1955) has shown that bacteria are able to decompose certain surface-active agents as a source of carbon.

Conclusions

It is highly problematical whether one will ever understand all of the bacterial activities in sediments. The few examples given here are merely a beginning of the understanding of the complex activities of bacteria in the surface sediments of shallow marine bays. The fact that it is recognized that bacterial activities are not uniform in sediment should stimulate the geochemist interested in diagenesis to learn more about the function of the bacteria and other micro-organisms.

Acknowledgements—The writer wishes to acknowledge the National Institutes of Health Research Project RG5885 for financial assistance to obtain the original data presented in this paper.

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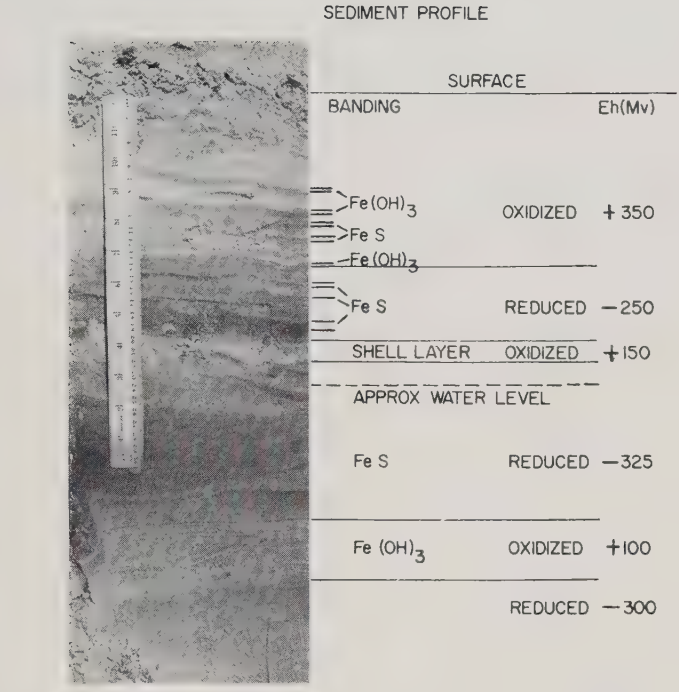
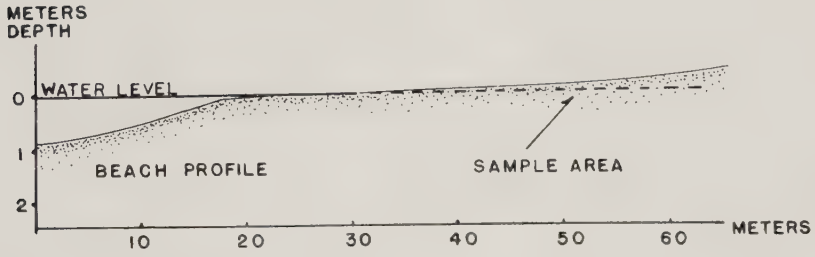


Fig. 8. Bacterial colour banding in surface sediments collected from along the edge of Redfish Bay, Texas.



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Some aspects of the distribution of biochemicals in geologic environments

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Abstract—Recent research on the distribution of biochemicals in soil is discussed in relation to the solution of problems in geochemistry. Consideration is given to the factors affecting the preservation of biochemicals in natural deposits. The results of a chromatographic study of the amino acids in soils are given. A new concept is presented concerning the nature of the nitrogen in sedimentary and igneous rocks.

Introduction

Remnants of plant and animal life, as well as products produced from them through metamorphosis, are preserved in ancient deposits. Included are fossils, sedimentary rocks, petroleum, coal, asphalt and "tar" sands. If the chemistry of the biochemicals in these deposits were known, there would be a better understanding of the origin and evolution of commercially-important products, such as coal and petroleum, and more knowledge of the early history of the earth.

Geologists are in general agreement that petroleum is of organic origin, yet the nature of the progenitors and the mechanism of their conversion into complex hydrocarbons remain obscure. It is also recognized that naturally-occurring organic compounds play an important role in weathering processes and in the geochemical cycle of certain elements (Breger, 1954; Schatz *et al.*, 1954), but practically nothing is known of the organic complexes involved. The suggestion has been made (Abelson, 1957, 1959; Miller, 1959) that biochemicals synthesized by ancient organisms, and by non-biological processes before life arose on the earth, are present in ancient deposits, but few attempts have been made to locate the deposits or to characterize their biochemicals.

The development of chromatographic methods of analysis has made it possible to isolate and identify minute amounts of specific organic compounds even when they occur in complex mixtures. However, before these procedures can be applied profitably to geologic specimens, the compounds must first be extracted and fractionated. The problem therefore becomes one of developing methods for the separation of organic constituents from inorganic matter without causing major changes in the chemical structure of the organic material. The interaction of organic compounds with inorganic constituents, particularly clay, deserves greater attention in the future. Bremner (1951) reviewed present information on the extraction of organic matter from soil and the interaction of organic and inorganic soil colloids.

The foregoing conclusions emphasize that there is ample justification for extensive research on the nature and properties of the organic chemicals in geologic environments. Unfortunately, the number of research workers in this field is small, and a great many problems remain unsolved. The solutions to many of the problems will require the combined efforts of scientists in several disciplines.

The formation of humus in soil has much in common with the initial stages in the formation of coal and petroleum. Information obtained by soil biochemists will therefore be of interest to petroleum and coal chemists. Since the biochemicals in soils, sediments and sedimentary rocks have been synthesized by organisms living during different periods of geologic time, a comparison of their biochemicals will provide valuable information about the comparative biochemistry of recent and ancient organisms.

Research in the Department of Agronomy at the University of Illinois (Stevenson, 1954, 1956a,b, 1957a,b, 1959a,b, 1960; Dhariwal and Stevenson, 1958; Stevenson and Dhariwal, 1959) has been concerned with studies of the distribution of organic constituents in soil and the chemistry of soil nitrogen. This review is designed specifically to cover the aspects of this work that may be of value to biogeochemists. A section entitled "Biogeochemistry of Nitrogen" will be of interest to all geologists.

Preservation of Biochemicals

Under anaerobic conditions, plant and animal tissues decay without extensive resynthesis by micro-organisms; under aerobic conditions even the most resistant plant parts are metabolized. Hence, aquatic sediments contain higher amounts of more or less modified plant and animal remains than do soils. Vallentyne (1957) has shown that chlorophyll-like compounds and carotenoids are extensively distributed in lake and marine deposits. They appear to be lacking in the humus of most soils, although Gorham (1959) detected chlorophyll derivative in the mull and mor humus layers of forest soils. Some of the biochemicals in sedimentary rocks may occur in fossils that are embedded or preserved in the rock deposits.

Biochemicals inherently resistant to attack by micro-organisms, such as lignin and lignin-like substances, tend to accumulate in soils and sediments. The combination of protein with lignin renders the protein less susceptible to decomposition. Humic acid, a dark-coloured colloid having many of the properties of oxidized lignin, may make up as much as a third of the organic matter in soils and sediments. Stevenson (1956b) observed that, in soil, this proportion decreases with depth in the profile. The salts of humic acid with di- and tri-valent cations are insoluble, a factor which undoubtedly increases their ability to persist in soil. The preponderance of humic acid in the organic matter of Chernozems, in contrast to that of podzolic soils, appears to be due to the fact that Chernozems contain larger amounts of calcium and other polyvalent cations. Dawson (1956) pointed out that dy muck is composed largely of insoluble salts of humic acid.

Support for the view that humic acids in soils and sediments are lignin-derived has been provided by the finding that humic acids have many of the properties of oxidized lignin. This evidence, although suggestive, is presumptive. Morrison (1958) found syringyl, guaiacyl, and *p*-hydroxyphenyl residues by alkaline-nitrobenzene oxidation of soil organic matter; however, the yields from mineral soil accounted for only from 0·5 to 1·0 per cent of the total organic carbon; from peat for only from 1·0 to 4·0 per cent. Gottlieb and Hendricks (1945) obtained good yields of aldehydic, acidic and neutral oils by both alkaline-nitrobenzene oxidation and high-pressure catalytic hydrogenation of soil organic matter, but were unable

to obtain characterizable products. The findings of Morrison, and Gottlieb and Hendricks, suggest that if soil humic acids are derived from lignin, the lignin has been drastically altered in the kinds and positions of the peripheral groupings on the aromatic rings.

Considerable attention has been given in recent years to the possibility that soil humic acids are products synthesized by, or formed by autolysis of, soil micro-organisms. Many micro-organisms, particularly the fungi and actinomycetes, produce dark-coloured humic-like substances. Brown nitrogenous polymers and copolymers also are formed by the condensation of amino compounds with substances containing an active carbonyl group, a reaction referred to as "browning". Hodge (1953) pointed out that browning can occur in the dry or nearly dry state at 25°C. The reactants required for browning are produced continuously in soils and sediments by autolysis of the bodies of micro-organisms, and the conditions in these deposits should be favourable for their condensation and polymerization. Stevenson (1960) suggested that the pigments in the fulvic acid fraction of soil organic matter were formed by browning processes. Bremner (1954), and Dawson (1956) discussed various theories regarding the origin of humic acids.

Very little is known of the genetic relationships that exist between humic acids in the various deposits. According to Waksman (1933), the humic acids of aquatic sediments are similar to those of terrestrial soils. Forsyth (1947a) suggested that all humic acids have similar molecular structures, but that the number and kind of reactive groups depend upon the conditions and length of time under which they were formed.

Forsyth (1947b) found that the fulvic fraction of soil organic matter could be separated into four parts by selective adsorption on activated charcoal. Fraction A, the effluent and washings (0·1 N HCl), consisted of water-soluble constituents, such as sugars and amino acids. Fraction B, obtained by elution with 90% acetone, was a pigment described as a phenolic glycoside. Fraction C, eluted with distilled water, was a polysaccharide containing glucose, galactose, mannose, arabinose, xylose and glucuronic acid (Forsyth, 1950). Fraction D, recovered with 0·5 N NaOH, was a dark-coloured material rich in nitrogen and containing pentose sugars and organic phosphorus compounds.

Stevenson (1960) found that Forsyth's fractions B and D were not single chemical entities, but were mixtures of several components. By the use of physico-chemical techniques, fraction B was separated into five components, four of which were pigmented. All of the fractions contained nitrogen which was liberated as amino acids by hydrolysis. Preliminary studies indicated that the pigments were not attacked readily by micro-organisms.

The origin and chemical composition of the pigments in soil are unknown. It would be of interest to determine if they are similar to those of other sediments, particularly those in deposits associated with petroleum formation.

Metallo-organic and organo-clay complexes

To some extent, the biochemicals in soils, sediments and sedimentary rocks are those which have been preserved selectively through their ability to combine with

metals and fine-grained mineral matter. Combinations are possible through complex formation with di- and tri-valent cations, and by adsorption on external surfaces and within the characteristic expansible layers of silicate minerals.

Evidence for the presence of metallo-organic complexes in soil has been given by Beckwith (1955), and Bremner *et al.* (1946). According to Bremner and his co-workers, the presence of the metals renders the organic matter in the complexes insoluble in water and in neutral solvents which do not themselves form complexes with the metals. The trivalent cations, iron and aluminium, form stronger complexes with organic matter than do divalent cations.

Breger (1954), and Schatz *et al.* (1954) pointed out that organic materials having chelating properties are important agents in weathering processes and in the geochemical cycle of certain elements. Soluble metallo-organic complexes migrate downward in soil, where they are protected somewhat from microbial attack, giving zones of organic matter accumulations. The faces of the fissures and cracks in the subsurface of many soil types are coated with dark-coloured organic matter. The translocated organic matter is high in ash, particularly iron and aluminium.

The vertical movement of organic matter, in combination with sesquioxides, is particularly pronounced in the development of the Podzol. The *B*-horizon of humus Podzols may contain from 10 to 20 per cent organic matter by weight. Kaurichev, *et al.* (1958) found that natural solutions of iron–organic complexes were highly stable; iron did not precipitate from some of their solutions upon standing for 1 year. They concluded that ferrous iron, formed in periods of temporarily excessive moisture, entered into interaction with water-soluble organic matter and formed complex iron–organic compounds, which upon subsequent oxidation of the iron retained great solubility and capacity to pass through soil. According to Stobbe and Wright (1959), products formed by the decomposition of organic matter in the surface soil bring about the solution of sesquioxides, the reduction of iron, and the formation of soluble organic complexes. The complexes precipitate in the lower horizons under oxidizing conditions and/or by sorption.

Many natural deposits, including crude oils, are known to contain dark-coloured organic materials which appear to be combined with metals. Wells and Erickson (1936) suggested the possible existence of calcium and magnesium salts of organic acids as constituents of petroleum, and as possible source materials for the formation of petroleum. Whether the organic materials in soils which complex with metals are similar to those in other deposits is a question that still needs to be answered. The trace elements in shales may have occurred originally as metallo-organic complexes.

Stevenson (1959b), and Bremner and Harada (1959) obtained evidence which indicated that as much as half of the organic matter in subsurface soil is held within the lattice structure of clay minerals. The interlayer compounds could not be extracted with alkali, but were recovered through the decomposition of the minerals with HF. Stevenson (1959b) suggested that organic matter has migrated in association with clay during soil profile development. Evidence in support of this hypothesis is provided by the observation of Buol and Hole (1959)

that the clay coatings on the peds of subsurface soil have considerably higher organic matter contents than the material in the interior of the peds. The biochemicals in aquatic muds, and in clay and marl deposits, may occur, in part, within the expandable layers of clay minerals.

The fixation of biochemicals within the lattice structure of silicate minerals is a highly selective process, and would be expected to result in the retention of biochemicals which would otherwise be lost to the sea by leaching, or destroyed by enzymatic or microbiological attack. The stability of biochemicals to ageing and

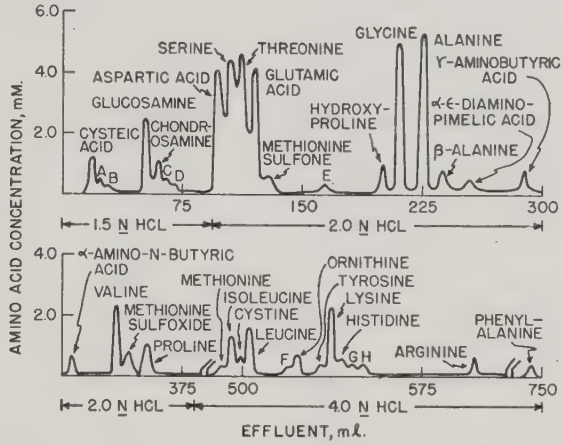

Fig. 1. Separation of ninhydrin-positive compounds from an acid hydrolysate of soil, using ion exchange chromatography. The resin was Dowex 50-X8 operated in the hydrogen form.

thermal destruction may be increased through their association with mineral matter, a factor which enhances the possibility of finding fossil biochemicals in an intact, or only slightly altered, condition. In certain situations, clay may act as a concentrating surface for substrate and exoenzymes, thereby bringing about a more rapid breakdown of the substrate, as suggested by Estermann and McLaren (1959).

Amino Acids in Soil

An analysis of the amino acids in soils, sediments and sedimentary rocks would permit a comparison of the nitrogen metabolism of organisms living under variable environmental conditions, and during different periods of the earth's history. Stimulated by the development of chromatographic techniques for detecting minute amounts of biochemicals in complex mixtures, the writer made a study of the amino compounds in soil hydrolysates (Stevenson 1954, 1956a, 1956b). A typical elution curve showing the separations obtained in a recent study using ion exchange chromatography is shown in Fig. 1. In all, thirty-six ninhydrin-reacting compounds were isolated. The compounds for which identification was considered positive are those listed on the diagram. The identification of N-acetyl-D-glucosamine, reported in the earlier study (Stevenson, 1956a) was not confirmed. This compound would be expected to be destroyed by hydrolysis. The compounds shown by the letters *E* and *F* have not been reported previously.

The identification of α-amino-*n*-butyric acid, γ-aminobutyric acid, and α, ε-diaminopimelic acid confirms an earlier finding by Bremner (1950). Young and Mortensen (1958) claimed to have detected fifty-seven amino compounds in soil hydrolysates, an observation which requires further study.

The presence of chondrosamine suggests that the amino sugars in soils are of microbial origin. The mucopolysaccharides of many micro-organisms contain chondrosamine, as well as glucosamine. Chitin, a compound found in the exoskeletons of certain lower animals, is a polysaccharide containing glucosamine units exclusively. A study of the relative amount of glucosamine to chondrosamine in soils, sediments and sedimentary rocks may prove of value in comparative biochemical studies. Apparently, no one has examined aquatic sediments or sedimentary rock for hexosamines.

The identification of α, ε-diaminopimelic acid and γ-aminobutyric acid in soil is of particular interest. The first-named amino acid appears to be confined to bacterial organisms, and is widely distributed among them. Gamma-aminobutyric acid, on the other hand, is known to occur extensively in plant and animal tissues, and has been detected in some micro-organisms. On the basis that the nitrogen compounds in soils are of microbial origin, it would seem that γ-aminobutyric acid may be more widespread than is generally recognized.

At present, it is not possible to distinguish between those amino compounds which occur naturally in soil and those which are artifacts producted by hydrolysis. Cysteic acid could arise from cystine, α-amino-*n*-butyric acid from threonine, ornithine from arginine, and methionine sulphone and methionine sulphoxide from methionine. Tryptophane is destroyed by acid hydrolysis, so its absence on the elution diagram was expected. It is of interest to know that Bremner (1950) was unable to detect tryptophane in alkaline hydrolysates of soil. Stevenson (1956a) obtained evidence which indicated that ornithine was a natural soil biochemical.

The compounds shown by the letters *F*, *G* and *H* on the elution diagram had low R_f values on paper chromatograms developed with collidine–lutidine as the solvent, and high R_f values with phenol as the solvent. These results suggest that the unknowns were basic in nature. Stevenson (1956a) was unable to detect *iso*lysine, methylhistidine, or hydroxy-lysine in soil hydrolysates.

Most of the amino acids shown on the elution diagram have been detected in hydrolysates of both the humic acid and fulvic acid fractions of soil organic matter. Bremner (1952) reported that the protein material of fulvic acids contains relatively large amounts of β-alanine and very small amounts of the basic amino acids, arginine and lysine. Both β-alanine and γ-aminobutyric acid were identified in the non-dialysable fraction of fulvic acid, indicating that they occurred in association with large polymers. Stevenson (1956b) found that cultivation brought about differences in the relative distribution of amino acids in soil.

It would be of interest to compare the amino acids of fossils and sedimentary rocks with those in present-day soils. Unfortunately, insufficient information is available on the amino acids in fossils and ancient sediments to come to any valid conclusions. Vallentyne (1957) gave a summary of the amino acids in hydrolysates of seston, water, fresh-water sediments, marine sediments and soil. Abelson (1957) identified alanine, glycine, leucine, valine, aspartic acid and glutamic acid

in a variety of fossil shells and bones. As indicated above, some of the amino acids in hydrolysates of soil and sediments could be artifacts; hence, the occurrence of a particular compound in a hydrolysate does not mean necessarily that the compound occurred naturally.

Biogeochemistry of Nitrogen

Nitrogen is distributed extensively throughout the silicate phase of the earth's crust. At present, very little is known of the chemical composition of this nitrogen. A review of the nitrogenous organic constituents in sediments has been given by

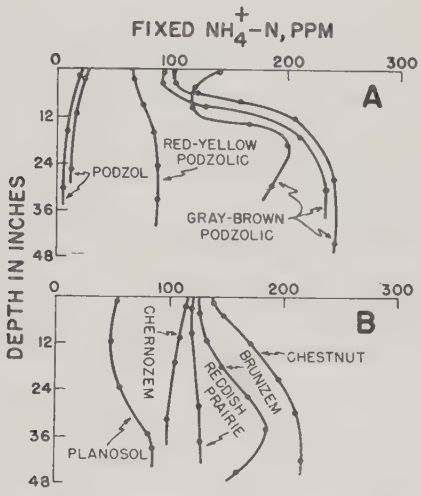

Fig. 2. Distribution of fixed ammonium in representative soils from several "great soil groups".

Abelson (1959). Bremner (1951, 1952) summarized the recent information on the nitrogen compounds in soil. Estimates of the geochemical distribution of nitrogen have been given by Hutchinson (1954).

For many years, soil scientists assumed that the nitrogen in soil occurred almost exclusively in organic combination; only a small fraction, generally of the order of 1 to 3 per cent, was considered to be in inorganic forms (as exchangeable ammonium and nitrates). Now it is known (Bremner and Harada, 1959; Dhariwal and Stevenson, 1958; Rodrigues, 1954; Stevenson and Dhariwal, 1959) that soils contain fixed ammonium (ammonium ions held within the lattice structure of silicate minerals). Stevenson (1959a) found that an average of two-thirds of the nitrogen in some Paleozoic shales was fixed ammonium.

The distribution of fixed ammonium nitrogen in representative soils of several great soil groups is shown in Fig. 2. All of the soils were from non-cultivated areas. The fixed ammonium was estimated from the amount of ammonium released from KOH-treated soils with HF, using the procedure of Dhariwal and Stevenson (1958). The results show that fixed ammonium was widely distributed in the soils of the various great soil groups. In some soils, the fixed ammonium increased with depth. The relatively low amount in the Podzol soils can be attributed to the low clay content of these soils.

The finding that sub-surface soils contain considerable quantities of fixed ammonium suggests that the parent materials from which the soils were formed contained fixed ammonium. A more extensive study with a Brunizem soil showed that fixed ammonium was distributed fairly uniformally to a depth of 82 in. Stevenson and Dhariwal (1959) suggested that some of the fixed ammonium in sub-surface soil existed originally as ammonia in the atmosphere, and became fixed by the minerals in early geological times as it was washed onto the earth's surface by rain water.

Carbon–nitrogen relationships in soils and sediments

The fact that fixed ammonium is widely distributed in argillaceous deposits affords an explanation for the low C/N ratios that have been reported for soils, sediments, and sedimentary rocks.

The C/N ratio of surface soil generally falls within well-defined limits, usually about 10 to 12 for the agriculturally important soils of the temperate regions. The ratio narrows sharply with depth; for sub-surface soil, reports of values below 5·0 are not uncommon. In view of the fact that the organic matter content of the soil diminishes sharply with depth, while the level of fixed ammonium remains fairly constant or increases, the deduction can be made that narrowing of the ratio with depth is due, at least in part, to fixed ammonium. Stevenson (1959b) discussed the relation between fixed ammonium and the C/N ratio in soil.

Hall and Miller (1908) determined the C/N ratios of some deep clays and marls and reported that several specimens had C/N ratios less than 5·5, and in one case a C/N ratio of 2·1 was reported. Mohamed (1949) found that the C/N ratios of some sediments from the northern Red Sea varied from 2·0 to 8·2 with a mean of 5·2. Arrhenius (1950) compiled the existing information on the carbon and nitrogen contents of sub-aquatic sediments and found that, in shelf sediments, the C/N ratio attained values between 4·3 and 12·7. In an earlier publication, Waksman (1933) summarized the information on the C/N ratios of marine sediments, and concluded that the ratios ranged from 5·3 to 13·3.

In view of the foregoing discussion, it is apparent that the C/N ratio of soils and sediments no longer can be considered a characteristic of the organic matter. Obviously, the results obtained by the Kjeldahl procedure no longer should be reported as "organic nitrogen".

Geochemical Distribution of Nitrogen

Since igneous rocks contain silicate minerals (micas, feldspars) which require the presence of appropriate cations to neutralize the negative charges arising from the substitution of aluminium for silicon in their lattice structures, it is reasonable to expect that they would contain fixed ammonium. This supposition has been confirmed by Stevenson (1959a), who found that the nitrogen in granite rocks was partly in the form of fixed ammonium. This reviewer suggests that the fixed ammonium in primary minerals was incorporated at the time the minerals were synthesized. Any deposit which contains minerals with silicate structures can be suspected of being an ammonium-burial trap.

Present information on the distribution of total nitrogen and fixed ammonium in sediments and igneous rocks is too fragmentary to permit an accurate estimate of their geochemical distribution. HUTCHINSON (1954) estimated the distribution of nitrogen to be as follows, where the values refer to the grammes of nitrogen corresponding to each square centimetre of the earth's crust.

	(g/cm_e^2)
In primary rock	32,000
In atmosphere	755
Fossil N associated with reduced C of sediments	67–108
N as nitrate in superficial deposits	2×10^{-5}

The value given by HUTCHINSON for the nitrogen of igneous rocks was based on data obtained by RAYLEIGH (1939), who found that the nitrogen content of igneous rocks averaged 0·005 per cent by weight. Results obtained in the writer's laboratory indicate that this value is high. The average nitrogen content of igneous rocks would appear to more nearly approximate a value of 0·0035 per cent by weight, but would probably occur within the range of 0·0025 and 0·005 per cent. Assuming that a third of the nitrogen in igneous rock (STEVENSON, 1959a), and half of that in sediments, occurs as fixed ammonium, the estimate for the distribution of nitrogen would be:

	(g/cm_e^2)
Organic N, ammonium salts, or exchangeable ammonium in primary rock	10,700–21,400
Fixed ammonium in primary rock	5,300–10,600
N in atmosphere	755
Organic N in sediments	34–54
Fixed ammonium in sediments	34–54
N as nitrate in superficial deposits	2×10^{-5}

The results given above show that the amount of nitrogen present in the earth's crust as fixed ammonium is of the order of from seven to twenty-two times that present in the atmosphere as elemental nitrogen. The suggestion of UREY (1952) that the nitrogen of igneous rocks is in the form of NH_4Cl clearly requires modification.

RUBEY (1951) concluded that the nitrogen in the atmosphere was generated from within the earth by volcanic activity during geologic times. This nitrogen could have arisen, in part, from the liberation of fixed ammonium through thermal destruction of the minerals. The stable form of nitrogen in the primitive earth could well have been as ammonium ions contained within silicate structures.

Acknowledgements—Appreciation is expressed to the Technical Committee of the North Central Cooperative Regional Project NC–17, and to the National Science Foundation for providing funds for this research.

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Geochimica et Cosmochimica Acta, 1960, Vol., 19 pp. 272 to 288. Pergamon Press Ltd. Printed in Northern Ireland

Petroleum pigments from Recent fresh-water sediments*

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Abstract—Pheophytin and other chlorin pigments are common in sediments deposited in fresh and marine environments due to the ready decomposition of the chlorophyll of the seston during deposition of the sediments. Laboratory investigations showed that it was possible to convert pheophytin to a petroleum-type porphyrin metal complex through a chlorin–metal intermediate. Examination of the dark organic gelatinous sediments of an almost stagnant autochthonous lake showed an appreciable content of trace hydrocarbons, in addition to chlorin pigments. Conversion of the chlorin pigments to petroleum-type porphyrins was apparent with increasing depth in the sediments, particularly when definite reducing conditions were established, at a position well below the top of the lake muds. Measurements of the rate of decomposition in the sediments carried out in the range 102–145°C and at pH 7·3, indicated the rapid decomposition of chlorins at lake-bottom temperatures. Measurements were also made of the rate of formation of nickel complexes of chlorins at 130–160°C, and at pH 3·6–7·3. In general, the rate of formation of the metal complexes was appreciably slower than the decomposition of the chlorins, especially when the availability of nickel was definitely limited. The instability of the chlorin pigments may account for the low pigment to hydrocarbon ratio of crude oils relative to that of the initial sediments. The rate studies and the observation of petroleum-type porphyrins in lake sediments indicate the formation of petroleum pigments very early in the development of crude oil.

Introduction

It is generally accepted that crude oil originates from sedimentary organic matter and there is little doubt about the occurrence of trace amounts of hydrocarbons and other potential crude oil materials in Recent fresh-water sediments (Judson and Murray, 1956) and marine sediments (Smith, 1954; Meinschein, 1959). It is also generally recognized that the porphyrins found in crude oils (Dunning *et al.*, 1954; Groennings, 1953; Hodgson and Baker, 1957) are derived almost solely from the original chlorophyll† (Treibs, 1936) in the source organic debris. In the study of the origin and accumulation of crude oil it is important to note the very close association of the hydrocarbon material and the pigmented substances from the source conditions through to the final reservoir rocks. Consequently it should be possible to establish a good understanding of the chemical conditions pertaining to the accumulation of crude oil from a knowledge of the chemical environments necessary for the obvious alteration of the original chlorophyll to the porphyrin pigments of crude oil

The trace pigments in crude oil are metal complexes of porphyrins and it is important to establish the manner in which they were formed from chlorophyll. Two major changes in the chlorophyll molecule are involved: one, in which the magnesium of the chlorophyll is replaced by a vanadyl, nickel or other metal ion (Baker and Hodgson, 1959), and the second, in which the chlorophyll molecule, which is a dihydroporphyrin molecule and not a porphyrin molecule, is altered to a porphyrin structure.

The alteration of the metallic constituent of the original chlorophyll could take place either by a direct replacement of the one metal, magnesium, by another, or

* Contribution no. 104 from the Research Council of Alberta, Edmonton, Canada.

† In the present paper the term chlorophyll is used to refer to chlorophyll *a* in general although it is recognized that in many instances minor amounts of other chlorophylls may be present with chlorophyll *a*.

it could result from a two-step process in which the magnesium is released forming a free chlorin pigment which is later re-complexed with another metal. Evidently the latter process has been predominant in Recent sediments. Chlorophyll *a* has not been observed in recent sediments although chlorophyll *b* has been reported (Vallentyne, 1959). On the other hand, pheophytin, formed from chlorophyll upon the release of magnesium, is abundant (Orr *et al.* 1958; Vallentyne and Craston, 1957). These findings are in good agreement with the demonstration of the very ready conversion of chlorophyll to pheophytin under simulated sedimentary conditions (Hodgson and Hitchon, 1959), and the observations made by Joslyn and Mackinney (1938) and Lamort (1956a, 1956b), who noted the relatively mild conditions under which chlorophyll is converted to pheophytin in homogeneous systems in the laboratory.

The conversion of chlorophyll which is a chlorin, a dihydroporphyrin, to a porphyrin pigment can be accomplished by a reduction of both the vinyl group, on position 2 of the basic structure, and the carbonyl group at position 9 in the isocyclic ring linking position 6 with the γ-carbon. The reduction of these two groups results in a spontaneous dehydrogenation at positions 7 and 8 thus converting the chlorin to a porphyrin pigment by the establishment of complete conjugation through a double bond linking carbons 7 and 8.

The reduction and dehydrogenation of chlorophyll may be brought about in many ways—both chemically and biologically. Willstäter and Stoll (1913) reported the conversion of chlorophyll and chlorophyllin by alcoholates to two monocarboxylic and two dicarboxylic acids, phylloporphyrin and pyrroporphyrin, and 2-vinyl rhodoporphyrin and rhodoporphyrin respectively, and these porphyrins can further be decarboxylated to give etioporphyrin (Karrer, 1947, p. 758). The conversion of the metal complexes of the chlorins to porphyrins is accompanied by a shift of the principal absorption maximum from the 640–680 mμ range characteristic of chlorophyll, to the 550–580 mμ range, characteristic of petroleum porphyrin complexes (Champlin and Dunning, 1958).

The path by which pheophytin is converted to vanadium and nickel complexes may involve one of two major intermediates as indicated in Fig. 1. In one, the pheophytin forms a metal–pheophytin complex, a chlorin complex, which is subsequently reduced and dehydrogenated to a metal-containing porphyrin. In the other, the pheophytin is reduced and dehydrogenated to a free porphyrin which subsequently complexes with a metal. Neither chlorin complexes nor free porphyrins have been reported in Recent sediments, but the apparent ease with which the nickel complex of pheophytin is formed (Baker and Hodgson, 1958) directed attention in the present investigation to the first possibility. Accordingly, a study was undertaken to establish the rate at which Recent sediment pigments could be converted to metal complexes under *in situ* sedimentary conditions.

Experimental

Conversion of chlorophyll to a nickel porphyrin

A study of the rate of formation of the metal–chlorin complexes could have little direct significance unless it was shown to be possible chemically to convert

chlorophyll to a petroleum-type metal–porphyrin complex by way of a metal–chlorin complex. Accordingly, the nickel complex of pheophytin was heated with 35% potassium hydroxide in ethylene glycol for about 30 min at 200–250°C, to produce a porphyrin-type material with absorption maxima at 390, 515 and 550 mμ, very similar to a nickel–porphyrin complex found in crude oil (Champlin and Dunning, 1958). The resulting pigment was established to be a nickel complex of a porphyrin by decomposing it to a free porphyrin with HBr (30%) in

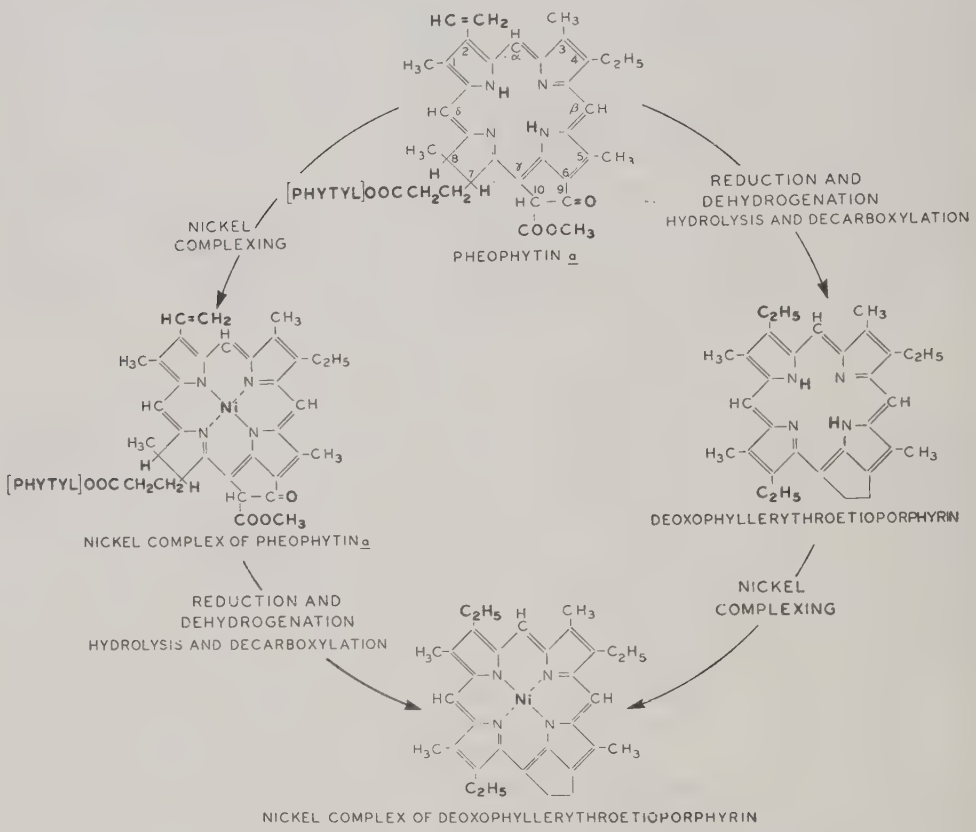

Fig. 1. Mechanisms of conversion of pheophytin *a* to a petroleum metal-complexed porphyrin.

acetic acid and observing a typical porphyrin spectrum (Rabinowitch, 1951), with absorption maxima at 400, 500, 530, 565 and 615 mμ—a spectrum very similar to that of deoxophyllerythroetioporphyrin (Sugihara and McGee, 1957). It was possible to react the free porphyrin further with nickel ammonium sulphate in acetic acid for a few minutes at 100–110°C to produce a metal–porphyrin complex very similar to the complex resulting from the reduction of the original nickel–pheophytin complex. In this manner it was established that the metal–chlorin structure could be converted by chemical means to a metal–porphyrin structure, even though the conditions were certainly more severe than would be expected in oil-bearing sediments.

Pigments in Recent sediments

The pigments in samples of Recent sediments from several lakes and rivers in Western Canada were extracted with 9 : 1 acetone–water mixtures. The absorption maxima for the extracts, listed in Table 1, show that pheophytin is a major constituent as indicated by major maxima at 418 and 667 mμ and the data agree well with the values reported by VALLENTYNE (1957) for sedimentary chlorophyll degradation products from muds of Connecticut lakes. The marine sediment

Table 1. Absorption maxima of pigment extracts from Recent sediments.

Source		Absorption Maximum (mμ)									
		400–500 (mμ)				500–600 (mμ)		600–800 (mμ)			
North Cooking Lake	22 miles E.S.E. of Edmonton, Alberta	*414**	448		480		535	600	*663*	690	740
Sylvan Lake	100 miles S. of Edmonton, Alberta	*414**	421	458	482		532	615	*666*	690	
Bidulock Lake	80 miles E. of Edmonton, Alberta	*412**	455		478		532	610	*664*		750
Five-mile Lake	5 miles N.E. of Mayo, Yukon	*408**	450		478		532	605	*660*		750
North Saskatchewan River	5 miles W. of Edmonton, Alberta	*406*†	432		478	508	530	605	*666*	680	750
Bonanza Creek	5 miles S.E. of Dawson, Yukon	410†	430		466	505	530	606	*666*		750
Laguna Madre	Texas, Gulf of Mexico	*415*‡	455		480		532	607	*666*		

Values in italics are principal absorption maxima.
* In acetone.
† In *iso*octane.
‡ In ether (HODGSON and HITCHON, 1959).

pigments reported by ORR *et al.* (1958) from the California coast and a Gulf of Mexico sediment pigment (HODGSON and HITCHON, 1959) are not significantly different from the fresh-water pigments. It is obvious that while there are minor variations, pheophytin-like Recent sediment pigments are ubiquitous and are generally available for association and accumulation with developing crude oils in non-marine as well as in marine sediments. One of the lakes examined in the present investigations—an almost stagnant lake characterized by a substantial thickness of lake-bottom gyttja—was selected for more detailed study. The lake chosen was North Cooking Lake, a shallow autochthonous lake about 22 miles ESE of Edmonton, Alberta.

North Cooking Lake

The lake basin occupies a depression in the Cooking Lake dead-ice moraine, and the till surface lies about 10 m below the present lake level at one sampling point.

The area of the lake is 3·1 miles^2 and the maximum depth of water was 2 m at a point about $\frac{1}{2}$ mile from shore. Beach sands, heavily contaminated with black organic matter, and rush-covered mudflats are predominant along the shoreline. Farther out in the lake up to 3·4 m of gyttja, underlain by 0·6 m of blue-black clay and 3·6 m of water-laid silts and sands were found above the till surface. A strong odour of hydrogen sulphide was associated with both the water and sediments.

Table 2. Variations of pH and redox potential in water and sediments in North Cooking Lake, Alberta.

Sample point	Thickness of sequence augered (m)	Depth below lake water surface (m)	Sample	pH	Redox potential (mV)
1	1·1 water and ice	0·9	water	8·1	+219
	3·7 gyttja	1·2	gyttja	7·4	+206
	0·6 blue clay	6·9	sand	8·6	+135
	4·3 sands and silts	7·6	sand	8·1	+119
	0·6 till (t.d.)	9·0	sand	7·6	+ 49
		9·3	till	6·7	+ 57
2	1·0 water and ice	0·9	water	7·1	+325
	0·5 gyttja	1·5	gyttja	7·4	+340
3	1·2 water and ice	0·5	water	6·5	+215
	0·2 gyttja	1·1	water	7·2	+220
		1·3	gyttja	7·2	+290
4	2·1 water and ice	1·1	water	8·1	+245
		1·5	water	7·9	+220
	1·2 gyttja	2·0	water	7·7	+245
		2·3	gyttja	7·2	+310
		3·4	gyttja	7·1	+335
5	2·4 water and ice	0·9	water	8·1	+225
	0·7 gyttja	1·6	water	7·6	+233
		2·4	gyttja	6·7	+244
		3·0	gyttja	6·8	+238
6	1·3 water and ice	1·2	water	7·8	+305
	2·1 gyttja	1·8	gyttja	7·1	+305
		3·0	gyttja	7·0	+310

Late in the winter (March, 1959) a field survey was made of the variations in pH and redox potential of the lake water and upper sediments by sampling through holes cut in the ice at six widely scattered locations. Portable pH and redox equipment was used to obtain immediate readings in the field so that negligible error could result from escaping hydrogen sulphide gas. The results, shown in Table 2, indicate a fairly high pH (about 8) in the water immediately under the ice cover, with a decrease to about 7·0–7·5 in the water just above the sediments and to values in the range of 6·7–7·4 in the upper sediments. The redox potentials of the water and upper muds were between +200 and +340 mV (relative to a

saturated calomel cell) as measured by a bright platinum electrode. These values fall well within the range reported by MORTIMER (1949) for the reversible transition of ferric to ferrous iron in limnological systems in which +200 mV was given as the boundary between oxidizing and reducing conditions. The porosity of the sediments was high, with values in excess of 90 per cent for some of the sediments, as shown in Table 3. Particle-size analysis of the gyttja indicated less than 5 per cent

Table 3. Variations of porosity and pH in near-shore sediments from North Cooking Lake, Alberta.

Sample point	Thickness of sequence augered (m)	Sample	pH	Porosity (%)
7	0·1 water and ice	silty gyttja, top	7·18	53·1
	1·0 silty gyttja	silty gyttja, middle	7·14	39·3
		silty gyttja, bottom	—	40·7
8	1·2 water and ice	gyttja, top	7·19	90·3
	2·4 gyttja	gyttja, middle	7·18	90·5
			7·17	91·8
	0·2 blue clay	gyttja, bottom	7·23	90·9
		blue clay	7·38	68·2
9	1·3 water and ice	gyttja, top	7·18	71·8
	1·3 gyttja	gyttja, middle	7·39	72·6
			7·28	67·9
		gyttja, bottom	7·68	70·1
10	1·2 water and ice	gyttja, top	7·27	85·5
	1·4 gyttja	gyttja, middle	7·23	83·2
			7·35	66·6
		gyttja, bottom	7·66	68·5

greater than 60 μ (mostly ostracode shells and fine silt), 20 per cent between 60 and 10 μ, 25 per cent between 10 and 3 μ, 20 per cent between 3 and 1 μ and 30 per cent less than 1 μ. X-ray diffraction analysis of the less than 1 μ fraction showed it to be composed largely of chlorite, probably derived from the till, with lesser amounts of illite, quartz and carbonates.

At one location about 200 yd from the shore, a more intensive study of the lake sediments was undertaken using a small rotary drilling rig supported by the lake ice to obtain Shelby tube samples of the lower lake sediments and of the underlying till. At this location there were 1·3 m of water, 3·3 m of gyttja, 0·4 m of dense blue clay and 3·3 m of sands and silts overlying the till. The pH and redox potential of the water were 8·1 and +219 mV; of the upper gyttja, 7·4 and +206 mV; of the sands and silts, 8·6 to 7·6 and +135 to +49 mV; and of the till 7·5 to 6·7 and +86 to +57 mV, respectively. Some iron staining, rare coalified woody fragments and gypsum spheroids up to 1 in. in diameter were observed in the lower 2 to 3 m of the silts. Some sections of the silts were well laminated.

The basal 1·5 m of sands and silts overlying the till are unfossiliferous. The

first elements of the ostracode fauna to colonize the lake were *Candona marchica* Hartwig, *Candona* "aff. *Cypris pubera* Müller" (Swain, 1947), and a species of *Limnocythere*. *Cyclocypris forbesi* Sharpe and *Potamocypris smaragdina* Vavra, both inhabitants of permanent quiet waters, make their appearance in the upper part of the sands. These latter two species are present in the overlying blue clay and in the gyttja, and *Potamocypris smaragdina* Vavra is an important element of living faunas. The clay fauna, which includes four species of *Candona* and three species of *Limnocythere*, is the most diversified as regards ostracode species; thus ecologic conditions were probably at an optimum at the time of blue clay deposition. Faunas become more restricted in the overlying gyttja—*Limnocythere* is absent from present faunas and *Cyclocypris forbesi* is rare. Species of *Candona* dominate the faunas throughout. Species of *Pisidium* are found in varying abundance in the gyttja, and present faunas include two species of fresh-water gastropods.

A preliminary microbiological examination of the sediments indicated a greater abundance of micro-organisms in the surface gyttja than in the lower sediments. A simple nutrient agar plate count showed about 2500 viable aerobic and facultatively anaerobic organisms (mainly spore formers) per gramme of the wet material. While contamination of the lower samples by surface material could not definitely be excluded, similar tests showed that at a depth of 1·8 m the count had fallen to about 500 organisms per gramme and in the silts at a depth of 3·6 m very few micro-organisms were found. After 3–4 months incubation there was some indication of microbiological sulphate reduction in media inoculated with surface gyttja and silt from 1·8 and 3·6 m in depth. No evidence of sulphate reduction was obtained from deeper samples.

The salinity of North Cooking Lake water at 1·83 p.p.t. indicates a considerable degree of stagnation and evaporation when compared with the average salinity of lake and river waters of 0·146 p.p.t. (Hutchinson, 1957, p. 553). Other significant features evident in Table 4 are the predominance of sulphate over chloride and the high content of hydrogen sulphide in the water below the ice.

The carbon content of the sediments from North Cooking Lake was made up for the most part of organic carbon with less than one-third being accounted for by carbonate carbon. Total carbon content of the gyttja was about 18 per cent, dry basis, with less than 1 per cent carbon occurring in the underlying silts. The soluble organic matter was obtained by exhaustively extracting samples of wet sediment first with acetone and then with benzene–methanol (10:1). The extract was reduced to dryness, the residue extracted with *n*-heptane and the resulting solution chromatographed on alumina (alumina adsorption, 80–200 mm, Fisher Scientific Company) to obtain paraffins-plus-naphthenes, aromatics, oxygen–nitrogen–sulphur compounds and residue, by elution with *n*-heptane, benzene and a mixture of acetone, methanol and pyridine, respectively. The results for five samples of sediment, corrected for solvent blank values and rounded off to two significant figures, are summarized in Table 5. It is apparent that very appreciable amounts of hydrocarbon materials were present, particularly in the gyttja. As would be expected, the hydrocarbons were much less abundant in the blue clay and in the silts. The hydrocarbon content of the two lower gyttja samples from North Cooking Lake was similar to the content of hydrocarbons in the gyttja and

Table 4. Water analysis, North Cooking Lake, Alberta*
(sample point 1)

	(mg/l.)
Cl	31
CO_3	0
HCO_3	859
SO_4	458
OH	0
Ca	98
Mg	62
Na	350
K	53
H_2S	45
total solids (at 180°C)	1400
non-ignitable suspended matter	0·7
V (p.p.m.)	0·002
Ni (p.p.m.)	0·002
density, 60°F	1·002
pH	7·7
refractive index, 25°C	1·3326

* All determinations except suspended matter, sodium, potassium, hydrogen sulphide, vanadium and nickel, courtesy Oil and Gas Conservation Board, Edmonton, Alberta.

Table 5. Organic content of North Cooking Lake sediments
(sample point 1)

	Gyttja (top)	Gyttja (middle)	Gyttja (bottom)	Blue Clay	Sand
total carbon	180000	180000	190000	42000	6300
organic carbon	130000	—	—	—	—
extractable organic matter	19000	16000	17000	3100	340
portion of extract soluble in *n*-heptane	9500	9300	7900	1400	160
(1) paraffins plus naphthenes	290	130	80	40	10
(2) aromatics	660	130	180	30	5
(3) O–N–S compounds	3800	2500	3400	600	70
(4) retained on alumina	4800	6500	4300	650	75
$\frac{\text{aromatics}}{\text{paraffins-plus-naphthenes}}$	2·3	1·0	2·3	0·75	0·5
$\frac{\text{O–N–S compounds}}{\text{paraffins-plus-naphthenes}}$	13·1	19·2	42·5	15·0	7·0

All values p.p.m. dry weight basis.

buff marl reported from Wisconsin lakes by Judson and Murray (1956), though higher than those reported from marine sediments off California (Orr and Emery, 1956) and considerably higher than those found in the Gulf of Mexico marine sediments (Stevens *et al.*, 1956). The surface gyttja from North Cooking Lake is remarkably enriched in both paraffins-plus-naphthenes and aromatics.

The method of extracting pigments quantitatively from the North Cooking Lake sediments was similar to that used by Orr *et al.* (1958) and by Vallentyne (1955). Samples of wet sediment weighing about 20 g were extracted at least three times with 40 ml of acetone containing 10 per cent water. In the extraction process the suspension was shaken for 5 min, centrifuged and decanted. Following the final extraction the residue was dried and weighed.

The gyttja is characterized by a pigment content of about 300 p.p.m. of the dried sediments assuming the pigments has extinction coefficients at about

Table 6. Pigment content of North Cooking Lake sediments

Sampling point	10	9	8
Distance from shore (m)	15	70	225
Thickness of samples section (m)	1·4	1·3	2·6
Pigment concentration (p.p.m. dry wt. basis)			
Sample (1) gyttja	300	280	350
(2) gyttja	250	140	310
(3) gyttja		72	360
(4) gyttja		78	280
(5) blue clay			110
(6) silts			2

667 mμ equal to that of pheophytin *a*. For six lake sediments Vallentyne (1955) reported somewhat higher values, up to about five times as high. Orr *et al.* (1958) found averaged pheophytin contents ranging from 0·05 to 100 p.p.m. in marine sediments from the California coast. The data for North Cooking Lake, shown in Table 6, therefore appear to be intermediate in magnitude. Both the values reported by Vallentyne (1955) and by Orr *et al.* (1958) and those for North Cooking Lake are consistent in that high pigment contents are in the upper sediments and gradually fall concomitantly with depth, except where a change in sediment type takes place. This is particularly apparent for the pigment contents of the arenaceous beds from North Cooking Lake.

The content of pigments in the major portion of the North Cooking Lake gyttja is of about the same magnitude as that of the combined paraffins, naphthenes and aromatics. The pigments are found only in the O–N–S fraction, however, where they comprise but a minor amount.

The composition of the pigment aggregates obtained from the majority of the samples analysed appeared to be similar both areally across the lake and vertically in a particular hole, although the total amount of pigment varies vertically in the sediments, as noted above. The total pigment aggregate from the middle of the gyttja bed was extracted and chromatographed on cellulose fibre using *iso*octane. Pheophytin *a* was one of the major pigments present and the aggregate was appreciably more complex than similar extracts described by Vallentyne (1955). The absorption spectra for ten of the most abundant pigments so recovered are listed in Table 7. The seventh pigment listed was indistinguishable from pheophytin *a* prepared from chlorophyll *a* by the method of Livingstone *et al.* (1953).

While the majority of the pigment extracts were very similar, a strikingly

significant change in pigment character was noted in the deep hole which penetrated to the till. Below the blue clay, the silts contained a coloured substance which was apparently a single metal–porphyrin complex. Although the pigment content of the silts was very low, about 2 p.p.m., thus making identification of the coloured material very difficult, there was little doubt about the general identification of it as a metal-complexed porphyrin. The extract from the silts was chromatographed on cellulose fibre using *iso*octane and the pigment was separated from some of the background components which had no specific absorption bands in the visible

Table 7. Absorption maxima of pigments extracted from North Cooking Lake gyttja.

Fraction number	Absorption maxima (mμ)			Solvent	Colour
	400–500 mμ	500–600 mμ	600–800 mμ		
1	*455*			*iso*octane	yellow
2	*480*			*iso*octane	pink
3	*420*	595	*655*	chloroform	grey
4	*415*		612, *670* 765	chloroform	grey
5	*418*, 460, 482		612, *670* 760	chloroform	yellow-pink
6	*430*, 458		628, *688* 765	chloroform	red-orange
7	*417*	505 535	615, *688*	chloroform	green-grey
8	*415*		612, *675*	chloroform	orange
9	*415*, 455, 483		610, *662* 762	chloroform	pink
10	*418*, 450, 480		610, *668*, 700, 765	chloroform	orange-beige

Values in italics are principal absorption maxima.

region. Absorption maxima at 410, 536 and 576 mμ in chloroform were observed for the single porphyrin complex, thus its spectrum is very similar to that reported by Champlin and Dunning (1958) for the vanadium complex from the McMurray crude oil. The significant features of these observations are:

(1) In the silts the number of pigments present is very small, contrasting with the ten or more present in the gyttja.
(2) The pigment is a porphyrin and not a chlorin.
(3) The pigment is a metal complex rather than a free porphyrin.

It is very important to note that the appearance of the metal–porphyrin complex coincided with very low redox potentials in the cores (+57 to +135 mV), potentials well below the +200 mV reducing threshold.

Formation of metal complexes in North Cooking Lake sediments

Two minor investigations were undertaken prior to the major detailed study concerned with the rate of formation of metal complexes. One dealt with the thermal stability of the sediment pigments and the other with the relative ease of complexing the two most abundant trace metals in crude oils—vanadium and nickel (Hodgson, 1954)—with the sediment pigments.

The thermal stability of the sediment pigments was measured by heating evacuated, sealed glass tubes containing North Cooking Lake gyttja at temperatures of 145°C and 102°C for periods of time ranging from 0·5 to 24 hr and

extracting the contained pigments. Comparison of the "reacted" pigments with the initial pigment aggregate showed a more severe destruction of the components responsible for the 667 mμ. absorption maximum than those with an absorption maximum at 680 mμ. To obtain an indication of the stability of the total pigment aggregate at lake bottom temperatures, the date for "reacted" pigment content in Table 8 were extrapolated to those conditions. Because of the disparity in the rates of degradation for the various components of the aggregate the rate for the overall reaction was not exactly first order. Nevertheless, it is clear that the pigments

Table 8. Thermal degradation of gyttja pigments in the sediments

Temperature (°C)	Reaction time (hr)	Pigment content (p.p.m. dry wt. basis)
102	0	393, 363, 369, 372
102	12	183, 183
102	16	165, 159
102	20	144, 144
102	24	123, 132
145	0	387, 332, 400
145	0·5	306
145	1·0	190
145	2·0	120
145	4·0	75

could not persist very long under the conditions at present characteristic of the gyttja. Again it is pertinent to note that the gyttja redox potential is well above the 200 mV reducing threshold.

An initial series of metal-complexing reactions was undertaken to test the interaction of both vanadium and nickel with the sediment and its contained pigments. Vanadyl sulphate and nickel ammonium sulphate were added separately to the gyttja in excess of the stoichiometric amounts required for the complexing reactions. Elevated temperatures and increasing amounts of acetic acid were used to facilitate metal complexing. At a pH of about 4 and a temperature of about 150°C it was observed that a reaction other than partial destruction of the chlorin aggregate occurred only when nickel was present. Following such a reaction, the pigments were extracted in the usual manner with a 90% acetone–water mixture and chromatographed with *iso*octane on cellulose fibre. Although many coloured compounds were present it was obvious that substantial alterations had taken place as indicated not only by the shift in the absorption spectra for the major components of the pigment system, but also by the nickel contents of the extracted fractions. The alteration of the wavelength of the principal chlorin band from 667 mμ, characteristic of pheophytin and its related starting materials, to 640–655 mμ, characteristic of the nickel complex of pheophytin (Baker and Hodgson, 1958), in the third, fourth, fifth and sixth components of the reacted material listed in Table 9 was good evidence of the formation of nickel–chlorin complexes. This conclusion was confirmed by the sharp increase in the nickel concentration for the

b and *c* fractions of Table 10, corresponding approximately to the third to sixth components of Table 9.

After demonstration of the ready formation of nickel complexes in the lake sediments under examination, a comprehensive study was undertaken of the rates of formation of such complexes as a function of pH and temperature in the ranges of 7·3 to 3·6 and 129 to 161°C, respectively. Gyttja from North Cooking Lake was prepared for the complexing reaction by the addition of nickel ammonium sulphate, to give a large excess (150-fold) of nickel, and acetic acid, to give the desired pH.

Table 9. Absorption maxima of pigments extracted from reacted North Cooking Lake gyttja.

Fraction number	Absorption maxima (mμ)*			Solvent	Colour
	400–500 mμ	500–600 mμ	600–800 mμ		
1	462			chloroform	pink
2	*415*		610, *667*, *762*	chloroform	yellow-green
3	422, *450*		605, *655*, 762	chloroform	beige
4	*420*		610, *655*, *763*	chloroform	beige
5	*420*		605, *650*, 762	chloroform	beige-green
6	*415*	595	*640*	*iso*octane	—
7	*412*		615, *670*, 755	*iso*octane	—

* Absorption maxima for nickel complex of pheophytin are *413*, 600 and *647* (Baker and Hodgson, 1958).

Values in italics are principal absorption maxima.

The gyttja so treated was transferred to thick-walled glass reaction tubes and the tubes were frozen, evacuated and sealed. A series of four or five identical tubes was used to establish the "half-time" of the complexing reaction for a given set of conditions. It was recognized that many reactions were involved for the many components present and that the interpretation of the overall results would be difficult. Nevertheless there appeared to be two main reactions: one in which the pigments were destroyed and the other in which the complexing took place. Accordingly, the "half-time" of the reaction was defined as the time at which the absorption peak representing "chlorin remaining" was equivalent in magnitude to the peak representing "complex formed" as illustrated in Fig. 2.

The data for the "half-time" of formation of the nickel complexes at four temperatures and four pH values, shown in Fig. 3, make it clear that the reaction between sediment pigments and nickel should proceed under the conditions obtaining in the sediments. The effect of pH does not seem to be particularly significant except under quite acid conditions, where the temperature dependence of the reaction is much more marked. At temperatures as low as 10°C only a few decades would be required for the formation of significant amounts of nickel complexes, provided that adequate supplies of nickel were available. In the experimental study a considerable excess of nickel was available, although in sediments this is not likely to be the case. In North Cooking Lake the nickel content of the water is only 0·002 p.p.m. If it is assumed that the interstitial water in the gyttja also contained 0·002 p.p.m. nickel, the molar ratio of nickel to chlorin

would only be about 0·01 to 1. The amount of available nickel from the sediments is still probably very small even though the total nickel content of the sediments is probably very much higher than that of the water, as indicated by Rankama and

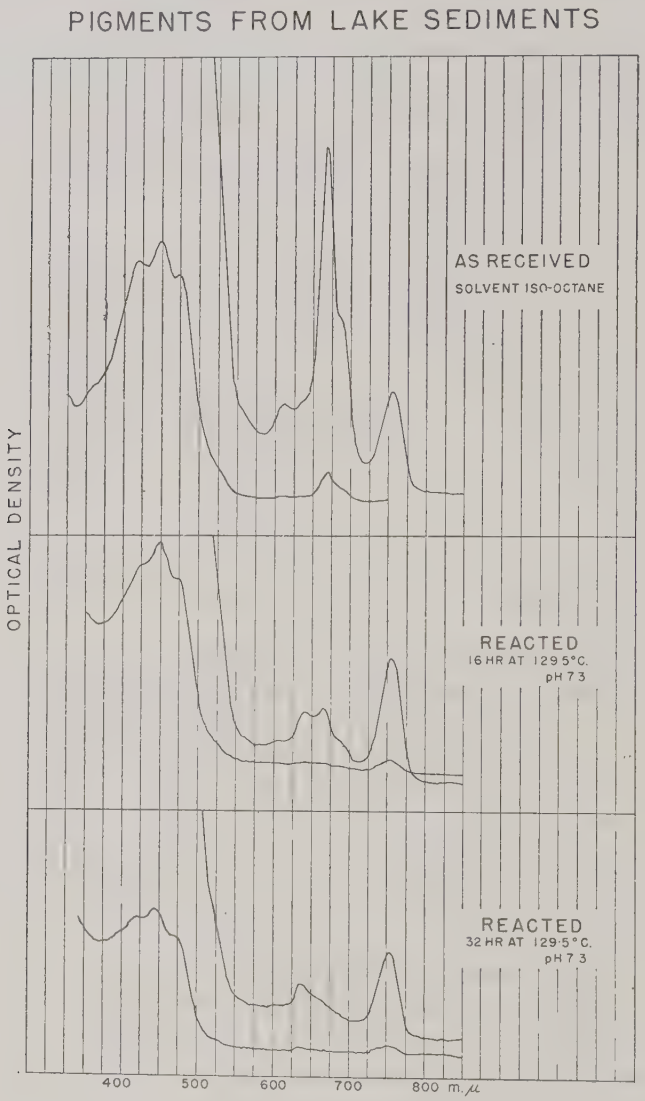

Fig. 2. Spectra of pigments from lake sediments illustrating original, "half-time" and "totally complexed" pigments.

Sahama (1950, p. 226) in summarizing nickel contents of sandstones and shales at 2–8 and 24 p.p.m., respectively. It is not known whether nickel is concentrated in fresh-water plants, but Mitchell (1948) and Young and Langille (1958) have summarized the contents of nickel in land grasses (0·5–4·0 p.p.m.) and marine algae (0·3–210 p.p.m.), respectively. It is probable that fresh-water plants

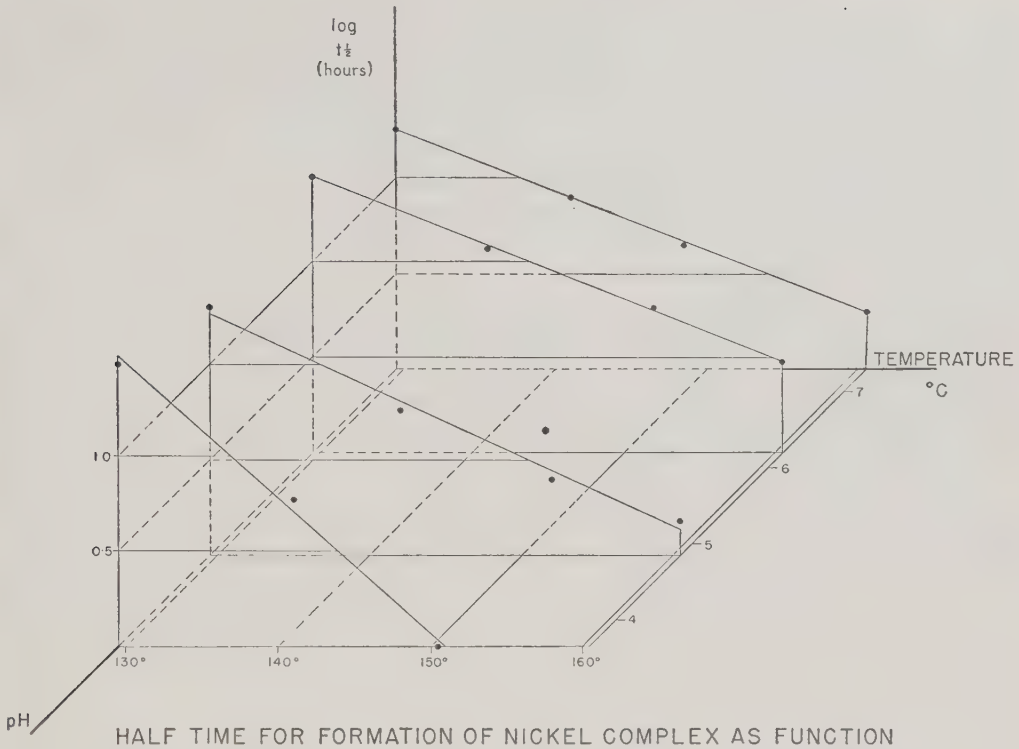

Fig. 3. "Half-time" for formation of nickel complex as a function of temperature and pH.

Table 10. Nickel content of pigment fractions from original and reacted North Cooking Lake gyttja.

Fraction	Nickel content (p.p.m.) of pigment extract	
	Original gyttja	Reacted gyttja
a	50	30
b	300	1600
c	500	1300
d	600	600

concentrate nickel in a similar manner. Decomposing plants could therefore supply appreciable quantities of soluble nickel salts to the interstitial water in the sediments. It seems reasonable to conclude that the nickel present in the sediments may react to a marked extent with the pigment aggregate.

Discussion

In the matter of the production of petroleum porphyrins from Recent fresh-water sediments, fundamental concepts have been explored and related to *in situ*

observations. Chemically it was shown that it is possible to convert pheophytin, the first decomposition product of chlorophyll, to a petroleum porphyrin pigment by way of a nickel complex of pheophytin. The other probable way of producing a petroleum porphyrin, by way of a free porphyrin intermediate, was not explored in detail although preliminary results indicated its feasibility. Examination of North Cooking Lake post-glacial lake sediments revealed the apparent conversion, at depth, of the many chlorin pigments characteristic of recent sediments, to a single metal-complex of a porphyrin which was evidently identical with the major porphyrin of most crude oils.

Laboratory studies of the rate of decomposition of chlorins demonstrated the instability of these pigments, while other rate studies showed the possibility of formation of nickel complexes of the pigments under sedimentary conditions, provided that adequate supplies of nickel were present. Thus the first stage of the overall conversion process which depends upon the production of a metal–chlorin intermediate was shown to be feasible under sediment conditions. The other process, involving a free porphyrin intermediate, is also compatible with the laboratory results and it may be responsible for the appearance of the vanadium rather than the nickel complex of the porphyrin found in the sediments, especially since the nickel complex of pheophytin is apparently formed more readily than the vanadium complex. Thus, although nickel apparently complexes with the chlorin more readily than vanadium the reverse may well be the case for the complexing of these metals with a free porphyrin. In summary, the feasibility of the first stage of the formation of petroleum pigments through a metal–chlorin intermediate was established, although this may not be the more important path.

Fundamental to either process for the formation of a petroleum pigment from the chlorins of the sediments, is the reduction of the carbonyl and vinyl groups of the chlorins, resulting in the spontaneous dehydrogenation of the pigment to form a porphyrin. In the laboratory very severe conditions were used to effect this change, and it is important to consider what conditions might develop in the sediments to bring about the same reactions. While microbiological agents are undoubtedly involved, and it is known that chlorins can be converted to porphyrins biologically (Vannotti, 1954, p. 57) it is pertinent to observe that only the sediments containing the porphyrin–metal complexes were characterized by reducing potentials. Although it appears, therefore, that porphyrins result from sediment processes requiring both considerable time, as indicated by the position of the porphyrin-containing sediments just above the till, and reducing conditions, characteristic of the silts overlying the till, it cannot be overlooked that lake conditions have not been static since glacial time and consequently other factors may have been operative at the time of the deposition of the silts. Faunal evidence shows the development of a fresh-water lake on the till surface with a considerable delay in the development of a population of organisms which could give rise to organic matter for the ultimate production of trace hydrocarbons and pigments. Optimum ecological conditions were reached only with the deposition of the blue clay overlying the silts. From that time to the present, restricted ecologic conditions were operative in the lake. While the gyttja presently being deposited may appear more typical of petroleum source sediments because of its organic gelatinous nature,

the underlying silts cannot be overlooked as playing an important role in the formation of crude oil. Although it is very likely that the pigments found in the silts are indigenous to the silts the possibility of migration of the pigments from the overlying sediments cannot be dismissed.

Taking into consideration the relationship existing between the pigments and the associated paraffins, naphthenes and aromatics, the silts appear to occupy a position intermediate in the development of crude oil between gyttja and crude oil reservoir sediments. In the gyttja the paraffins, naphthenes and aromatics are only slightly more abundant than the pigments; in the silts the ratio is about seven to one, and in crude oils about 10,000 to 1. Such a change in ratio could result either from a gain in the hydrocarbon content or a loss in the pigment content. While the former is not impossible from a consideration of the thermodynamics of the conversion of organic debris to hydrocarbons, the latter is consistent with the measured thermal instabilities of the pigments. Although other factors such as differential solution and adsorption during migration of the developing crude oil may be important, both the possibilities of gain of hydrocarbons and loss of pigments are in agreement with the conclusion that the organic matter in the silts has been substantially altered toward a crude oil as indicated both by the sharply-reduced number of pigments and by the development of the porphyrin type of metal complex characteristic of crude oil.

Conclusions

Two general paths for the formation of petroleum pigments from lake sediment pigments have been outlined: one in which the major intermediate between the initial chlorin and final metal–porphyrin was a metal complex of the chlorin, and the other in which the major intermediate was a free porphyrin. Measurements of the rate of decomposition of the initial chlorin pigments in the sediment and the rate of reaction between pigments and soluble nickel established the possibility of the first alternative but did not rule out the second. The observation of the presence of metal complexes of porphyrins in the older deposits where reducing conditions had developed, while confirming neither path, did indicate that the formation of petroleum pigments takes place early in the development of crude oil, long before the oil is expelled from the compacting source sediments.

Acknowledgements—Grateful acknowledgement is given for the faunal and microbiological determinations made by R. Green and R. M. Macrae, respectively.

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Review of the chromatographic "plate" theory with reference to fluid flow in rocks and sediments

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Abstract—The classical theories of Martin and Synge are used to demonstrate the possibility of chromatographic flow in sedimentary rocks. A simple experiment is described which defines some of the limits of chromatography in quartz sand and in aqueous media.

Introduction

Chromatography is one of the principal methods of chemical analysis. In fact, chromatography is just as much of a part of fields of modern analysis as distillation is of organic chemistry or X-ray diffraction is of mineralogy. The theories of chromatographic separations have been sufficiently developed to enable one to evaluate and predict separation efficiency in many systems.

Keulemans (1957) gives the following definition: "chromatography is a physical method of separation" (of fluid mixtures), "in which the components to be separated are distributed between two phases, one of these phases constituting a stationary bed of large surface area, the other being a fluid that percolates through the stationary bed". The word "chromatography" refers to early experiments which involved the separation of coloured substances. The colour, of course, has nothing to do with the separation. In fact, the majority of substances now separated by chromatography are colourless.

Beds of many clastic sediments and also several fine-grained carbonate rocks have surface areas comparable to or even larger than the adsorbents used in the laboratory. They are also porous and through them, *in situ*, natural fluids percolate This has been known to students of petrography and geology for many years; consequently, speculations regarding the chromatographic character of the rock strata is not new. The term first used was "selective adsorption process". These speculations gained substance with the advancement of the theory and practice of chromatography.

It has been known since the early work of Day (1897) and Gilpin and Cram (1908) that selective adsorption occurs on passing crude petroleum through beds of some clay minerals. Light and low-boiling hydrocarbons come through first; higher boiling ones come later; asphaltic matter and some high molecular weight hydrocarbons are removed from the fluid phase. Mileshina *et al.* (1959) described experiments in which crude oil was passed through quartz sand and quartz sand, clay minerals, feldspars or calcite mixtures. Selective adsorption of non-hydrocarbon constituents was observed in addition to chemical changes in the crude oil filtrates. Field data suggesting geological chromatography became available when regional geochemical studies were undertaken. Bonham (1956) suggested that one of the possible causes of the variations in the trace element concentration of the crude oils in the Seminole area in Oklahoma "may be attributed to adsorption of

the polar, metal-bearing organic constituents by clay or other minerals in the rocks through which the oil migrated." Hodgson and Baker (1959) suggested that variations in the Ni and V content within several Alberta and Saskatchewan oil deposits might be connected with the fact that "the passage of crude oil through a porous solid may result in a retention of the high molecular weight and polar material." Maps showing the distribution of these trace elements within the oil fields were presented.

In a recent paper Brooks (1959) voiced the opinion that the occurrence of the asphalt-free crude oils of Pennsylvania probably resulted from a selective filtration process where the rocks acted as the chromatographic separating agents. Smith *et al.* (1959) raised the question that the absence or presence of certain compounds in petroleum may be the result of "natural chromatography".

In addition to these, it is generally known, that usually small quantities of a large variety of organic compounds are present in Recent marine sediments. These include hydrocarbons, such as those found by Smith (1954), O, N bearing compounds, and others. Petroleum and natural gas, which are the only major organic *and* fluid constituents of the sedimentary rock strata, show the predominance of hydrocarbons. The hydrocarbon content of a given reservoir rock indeed represents a preferential accumulation of sedimentary organic matter.

The origin and migration of petroleum is and has been a field of considerable research interest; this is probably the reason why geological chromatography has been mentioned mainly in connexion with petroleum problems. It must be, of course, a general phenomenon affecting all natural fluids flowing in rocks.

There are many indications that chromatography is operational in sedimentary rocks. The purpose of this paper is to attempt to prove the possibility of this in terms of classical theory.

The "Plate" Theory of Chromatographic Flow

Different physical and chemical processes effect the separation in the various chromatographic systems. The word "chromatography" is used as a collective term for a *group* of processes that at first sight may appear diverse. The processes, however, have certain common features; for example, they all involve the transport of a fluid mixture through a bed (or a column) of dispersed matter. The fluid mixture may be a liquid or a gas (vapour). The bed contains a substance, which is called the stationary phase, that consists either of a solid adsorbing agent or a liquid partitioning agent. The mixture is transported through the bed either by a gas or by a liquid. This is called the moving phase. The stationary phase selectively retards the transport of the components of the mixture in the moving phase; consequently, the components move through the bed at different effective rates. The separation is caused by the fact that the components of the mixture move as separate zones or "bands."

Such dissimilar causes as adsorption, differential solubility, ion exchange capacity and emulsion phenomena are responsible for the various chromatographic systems. Adsorption chromatography relies on the adsorptive distribution between a fluid mixture and the surface of a solid adsorbent. Differential solubility of the solutes of the moving phase in an essentially stationary, sorbed "liquid"

phase (usually a thin coating on an inert, solid support) is the cause of partition chromatography. (Gas chromatography, liquid-partition and paper chromatography fall into this category.) Ion exchange chromatography is effected by the ion distribution between a fluid mixture and the surface of the ion exchanger, stationary phase. Foam and emulsion chromatography is based on the distribution of the solute between the solution and the interfaces of immiscible liquid droplets moving through the solution. Chromatogaphy in sedimentary rocks may involve any one, or a combination of these four mechanisms.

It is possible to calculate the concentration of a solute at any place in the column. MARTIN and SYNGE (1941) postulated the now classical "plate-theory" of chromatography in analogy to counter-current extraction processes. According to the MARTIN and SYNGE concept the column may be subdivided into several hypothetical layers which are called the "theoretical plates." Within each of these layers equilibrium is reached between the moving and stationary phases. The height of one of these layers is called the h.e.t.p. or "height equivalent to one theoretical plate." The h.e.t.p. is defined as "the thickness of the layer such that the solution issuing from it is in equilibrium with the mean concentration of solute in the non-mobile phase throughout the layer."

Consider that a unit mass of a single solute is placed into the first plate of the column. This is followed by a pure solvent. After the passage of successive infinitesimal volumes of the mobile phase, δv, the quantities of the solute in the successive plates are as follows:

Volume of solvent passed $x\delta v$	Serial number of plates, r (measured from the top of the column downwards)				
	1	2	3	4	5
0	1	0	0	0	0
1	$(1\text{-}\delta v/V)$	$\delta v/V$	0	0	0
2	$(1\text{-}\delta v/V)^2$	$2(1\text{-}\delta v/V)\delta v/V$	$(\delta v/V)^2$	0	0
3	$(1\text{-}\delta v/V)^3$	$3(1\text{-}\delta v/V)^2(\delta v/V)$	$3(1\text{-}\delta v/V)(\delta v/V)^2$	$(\delta v/V)^3$	0
4	$(1\text{-}\delta v/V)^4$	$4(1\text{-}\delta v/V)^3(\delta v/V)$	$6(1\text{-}\delta v/V)^2(\delta v/V)^2$	$4(1\text{-}\delta v/V)(\delta v/V)^3$	$(\delta v/V)^4$

Consider the following symbols:

h = h.e.t.p;
A = cross-sectional area of the column;
A_S = cross-sectional area of the stationary phase;
A_L = cross-sectional area of the moving phase;
A_I = cross-sectional area of the inert solid ($A = A_S + A_L + A_I$);
v = volume of the solvent that passed through the column;
V_S = volume of the stationary phase in one theoretical plate;
V_L = volume of the moving phase in one theoretical plate;
$V = V_L + \alpha V_S = h(A_L + \alpha A_S)$ effective plate volume (dimensionless);
$v_r = v/V_L + \alpha V_S$;
r = serial number of plate;

α = partition (distribution) coefficient (i.e. the g solute per ml of stationary phase/g solute per ml of moving phase, at equilibrium);
R = distance of position of maximum concentration of solute/simultaneous distance of movement of the front of the solvent;
Q_r = total quantity of solute in plate r.

Because Q_r is a term of the binomial expansion $((1 - \delta v/V) + \delta v/V)^n$, after n successive volumes of solvent, δv, have passed;

$$Q_{r+1} = \frac{n!(1 - \delta v/V)^{n-r}(\delta v/V)^r}{r!(n - r)!}$$

When n is large,

$$Q_{r+1} = \frac{1}{r!}(n\delta v/V)^r e^{-n\delta v/V}$$

and because $n\delta v = v$,

$$Q_{r+1} = \frac{1}{r!}(v/V)^r e^{-v/V}$$

this becomes by Stirling's approximation:

$$Q_{r+1} = \frac{1}{\sqrt{(2\pi r)}}(v/rV)^r e^{r-v/V}$$

when r is large. And because $v_r = v/V_L + \alpha V_S = v/V$,

$$Q_r = \frac{1}{\sqrt{(2\pi r)}} e^{r-v_r}\left(\frac{v_r}{r}\right)^r \tag{1}$$

Q_r is listed instead of Q_{r+1}, because equation (1) may be used for the calculation of the quantity of solute issuing from the last plate of the column as well as for the calculation of the quantity of the solute in any plate preceding the last one.

The relative rate of movement of a band is expressed by

$$R = \frac{hv/V}{v/A} = \frac{Ah}{V} = \frac{A}{A_L + \alpha A_S}$$

therefore

$$\alpha = \frac{A}{RA_S} - \frac{A_L}{A_S} \tag{2}$$

The definition of α does not specify any particular mechanism for attaining distribution equilibria. Naturally, there is no reason why the partitioning effect would not take place in aqueous systems, such as in the formation fluids, provided the proper solute and adsorbent are present. Both (1) and (2) apply to the different systems of chromatography.

There are two assumptions in the theory that need mentioning. First the diffusion of the solute from one plate into another one is considered to be negligible. Second, the distribution coefficient, α, is assumed to be independent both of the absolute value of concentration and of the presence of other solutes. These assumptions are of course, not entirely correct regarding the laboratory columns. Neither can they be expected to hold true fully in the rock strata.

A Theoretical Proof of Geological Chromatography

Consider v_r which appears in equation (1),

$$v_r = \frac{v}{V_L + \alpha V_S}$$

It follows from (1) that chromatography does take place when

$$v_r > 1, \text{ when } r > 1$$

To the contrary, the fluid flow is unaffected by chromatography if, throughout the flow process, $v_r = 1$. It shall be shown that this condition may or may not be present in rocks.

It is evident that in order to maintain $v_r = 1$, throughout the flow process, the following condition has to be fulfilled:

$$v = V_L + \alpha V_S$$

which occurs when $\alpha = 0$ and then $V_L = v$.

It follows by definition that α has a value of zero only if there is no interaction between the fluid components and the mineral matter and/or the organic content of the rock. This occurs, for example, when certain solutes, such as non-polar compounds, flow through a coarse grained, "clean" quartz sand of low surface area.

When polar solutes are transported through rocks which contain colloidal sized mineral matter, such as the clay minerals, interaction between the solute and the rock must take place. Similarily, when the solute is soluble in organic inclusions present in the rock, interaction must take place. It is of general knowledge, of course, that a great many sedimentary rocks contain clays and/or organic inclusions. This necessitates that, $\alpha > 0$, when the appropriate solute is in contact, and, consequently, chromatography must be operational.

This elementary proof is offered to demonstrate that fluid flow in sedimentary rocks may be chromatographic in nature.

It may be added, that, because the fluids percolating through the rocks traverse great distances, appreciable separations could probably be accomplished on relatively inactive adsorbents (i.e. when α has a very small value).

A Laboratory Experiment in Geological Chromatography

Solutions of organic compounds in water were continuously introduced into quartz sand packed columns. In terms of the Martin and Synge theory this is equivalent to introducing successive unit masses of a solute into the first plate of the column. Chromatography is demonstrated if the solvent comes out first and the solute concentration builds up only after some pure solvent has accumulated in the effluent volume. If chromatography is absent the effluent concentration will equal the concentration in the input fluid throughout the entire flow process.

Medium grain size "Ottawa" quartz sand was fractionated by repeated decantations in water. A fraction was obtained that consisted of particles of uniform size; the median particle size was found to be 0·3 mm. Glass columns

were packed with the sand fraction by continued tapping. The columns were approximately 28 cm long; their outside diameter was 2·2 cm. Their porosity was approximately 50 per cent. The solution was introduced into the colums on the top from a reservoir placed approximately $\frac{1}{2}$ m above the top of the columns. The lower end of the columns discharged the effluent into vessels of known volumes. The columns were stored in a refrigerator in order to prevent contaminations by microorganisms.

Aqueous solutions were prepared (1) of the carbohydrate, glucose and (2) of the

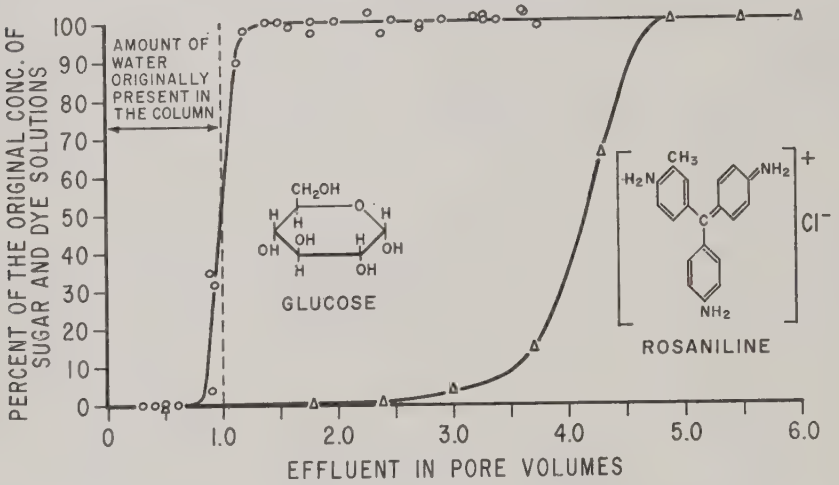
PERCENT OF THE ORIGINAL CONC. OF SUGAR AND DYE SOLUTIONS
AMOUNT OF WATER ORIGINALLY PRESENT IN THE COLUMN
GLUCOSE
ROSANILINE
EFFLUENT IN PORE VOLUMES

Fig. 1. Chromatographic effect in quartz sand columns.

triphenylmethane dye, rosaniline. The same sand fraction was used for both the glucose and the rosaniline experiments. The input concentrations of the two solutions varied; the concentration of the glucose solution was 0·1 M and the concentration of the rosaniline solution was 0·0001 M. The choice of the solutes and their concentrations were designed to enhance chromatographic or non-chromatographic flow in quartz sand. The triphenylmethane dye, rosaniline, does not, of course, occur in nature, and even glucose seems to have only a limited geological role. Nevertheless, they were selected for this experiment because (1) organic compounds of diverse character are known to enter the sediments and (2) because the ease of handling of these two compounds and previous experience with them in this laboratory, seemed to have assured maximum accuracy in the flow experiments. Only the rosaniline is strongly adsorbed from aqueous solutions on this fraction of quartz sand. The adsorptivity of rosaniline on sand has been measured by Nagy and Wourms (1959). The concentrations of the effluent sugar solutions were analysed by evaporating the water from the solution on a water bath and weighing the dry residue. The concentration of the dye, rosaniline, was determined colorimetrically with a Bausch and Lomb "Spectronic 20" colorimeter at 489 mμ wavelength.

Fig. 1 shows the results of the experiments. Because the columns were filled with water when the solutions were introduced, the solutions had to displace the

water from the pore space. The concentration of the effluent glucose solution built up to the level of the original concentration after approximately one pore volume of fluid had been displaced from the columns. This indicates that chromatography was absent when glucose passed through the sand.

The concentration of rosaniline effluent did not increase appreciably until approximately the four pore volume mark. The eluent came out first and the flow of the solute was retarded in the column. This shows that chromatography was operational.

The following conditions were indicated. (1) A common constituent of sedimentary rocks, quartz sand, may or may not act as a chromatographic adsorbing agent. (2) Quartz sand may induce chromatography in the presence of water, if (3) the solute is adsorbed on the sand grains from water.

No discussion of geological chromatography can be complete without mentioning the work of KOBAYASHI and his co-workers, even though the purpose of these researchers was not to simulate geological environments. They have found that partition chromatographic effects were operational during the flow of hydrocarbon gases through crude oil saturated rocks. ROPER *et al.* (1958) reported that methane, propane and helium travelled at different rates of flow through a 30 cm long Torpedo sandstone core containing 8·86 per cent Ventura crude oil. The results were expressed in terms of the retention time, which is the difference in time required for the non-adsorbed and the adsorbed components to flow through the column. The retention time of methane was found to be 0·18 min, that of propane was 0·85 min, and helium was not adsorbed in the core.

CONCLUSIONS

The classical, chromatographic plate theory of MARTIN and SYNGE points out that chromatography may be operational during the flow of mixtures of fluids through sedimentary rocks. Natural chromatography will be effective whenever the partition coefficient has a value greater than zero. The partition coefficient can have a value of zero only when the solute is (1) not adsorbed on the surfaces of mineral particles, (2) dissolved in organic matter in the rock, or (3) when it does not enter into ion exchange positions on clay minerals. The presence of clay and other colloidal-sized minerals and the presence of bituminous matter necessitates chromatographic flow of certain organic compounds.

Acknowledgements—The author wishes to acknowledge the support of the National Science Foundation in conducting research on geological chromatography at Fordham University. Messrs. R. F. ANGIOLELLI and R. RIECHERS assisted in conducting the flow experiments.

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Diagenesis of metabolites and a discussion of the origin of petroleum hydrocarbons

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Abstract—Proteins and carbohydrates are rapidly degraded to compounds of no direct interest in the problem of the origin of petroleum. Lignin, if carried into marine basins in the form of humic substances, is probably the major progenitor of kerogen rather than the precursor of petroleum. Pigments are but minor contributors to petroleum.

The fate of fatty acids in a marine environment is not completely understood. Although they may not be directly decarboxylated biochemically, it is shown how they can be converted into oxygenated or dehydrogenated acids more reactive than the parent compounds. Illustrations are also given for Diels–Alder reactions that could account for the formation from these compounds of the alicyclic and aromatic hydrocarbons in petroleum.

It is most likely that crude oil is generated in sediments containing concentrations of lipids, the character of which governs the nature of the oil that is formed.

There is no longer any argument regarding the organic origin of crude oil. Knowledge of the mechanisms whereby complex oxygen-, nitrogen-, and sulphur-bearing biological progenitors are converted into a complex mixture of hydrocarbons is, however, still limited. In an attempt to achieve a better basis for understanding how such conversion may have occurred, an effort has been made to consider the fate of various biological substances in diagenetic environments. In particular, it was of considerable interest to evaluate any interactions that could possibly occur between initial degradation products under the geochemical conditions of petroleum formation.

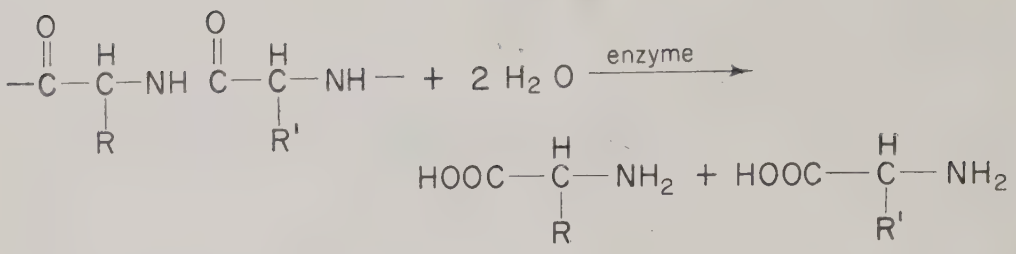
Crude oils are generally thought to be formed from complex organic materials in marine muds. Of necessity, initial consideration must be given to the nature of this organic matter in any study of the origin of petroleum.

Proteins, carbohydrates, and lipids make up the three major classes of substances produced in life processes; these three types of compounds along with lignin and pigments could, therefore, be expected to contribute to a marine sediment in which oil could later be formed.

Progenitors

Proteins

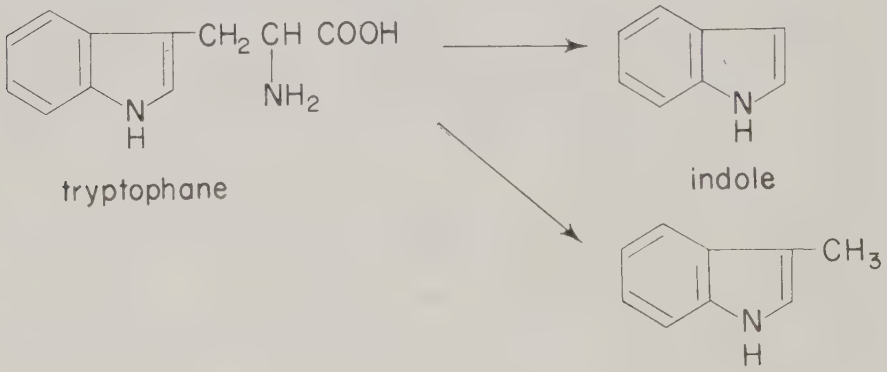
Proteins, both the water soluble and water insoluble varieties, are composed of fewer that thirty different α-amino acids tied together in peptide linkages. Most proteins are sensitive to attack by proteolytic enzymes, such as pepsin or trypsin, which hydrolyse the peptide bonds to form partially decomposed proteins. The latter are then attacked and degraded by other known enzymes. Proteolytic processes, which in effect hydrolyse the peptide bond, operate with but a slight energy requirement and serve to prepare the proteins for eventual utilization as energy sources on other oxidative or reductive biochemical processes. Although these processes and enzymes are those normally associated with human biochemical processes, there can be little doubt that analogous degradations occur in other animal systems. In general, these processes would be expected to result in

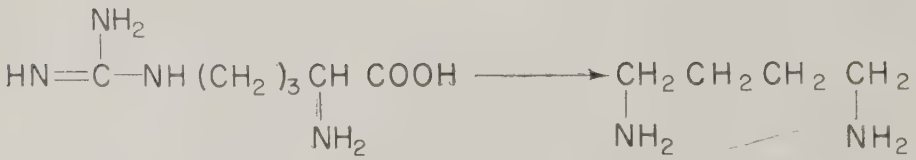


Hydrolysis of a peptide bond

the elimination of proteinaceous matter as carbon dioxide, water, ammonia, nitrogen and hydrogen sulphide. Other labile products, such as urea, would, if produced, eventually yield similar degradation products.

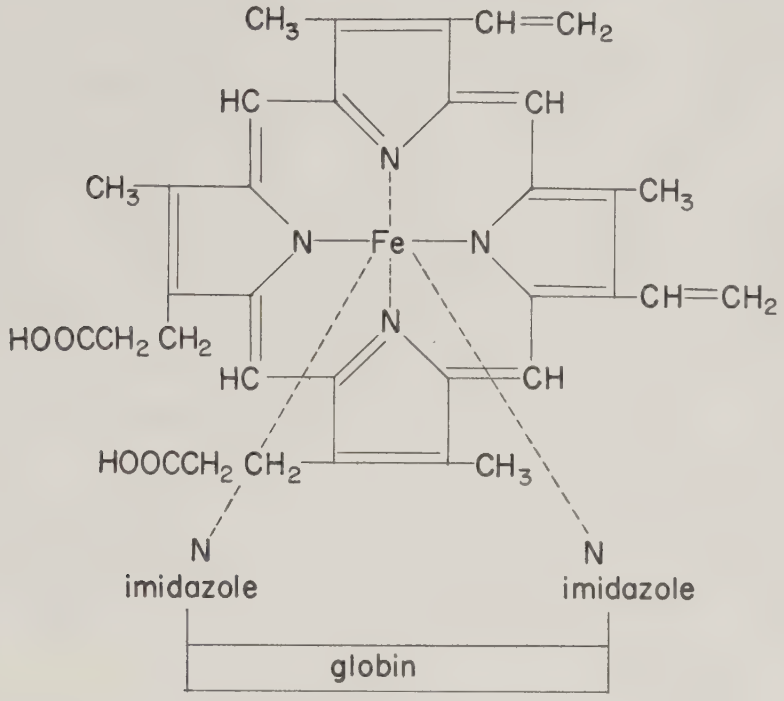
Proteins, therefore, appear to undergo biochemical degradations leading to compounds of no direct interest in the problem of the origin of petroleum. Only in certain cases of putrefaction can biological processes result in the formation of stable nitrogen bases that might be incorporated into sediments. Examples of these processes are the formation of indole and skatole from tryptophane, cadaverine from lysine, and putrescine from arginine.





Certain proteins of biological origin are associated with other compounds and are known as conjugated proteins. An example of such a compound is haemoglobin, in which the globin, a protein, is bound through imidazole nitrogen to protoporphyrin-III containing an atom of reduced iron (HARPER, 1957, p. 60).

It is evident that degradation of the protein will lead to a residue of porphyrin which, as evidenced by its occurrence in petroleum, is resistant to biochemical decomposition.



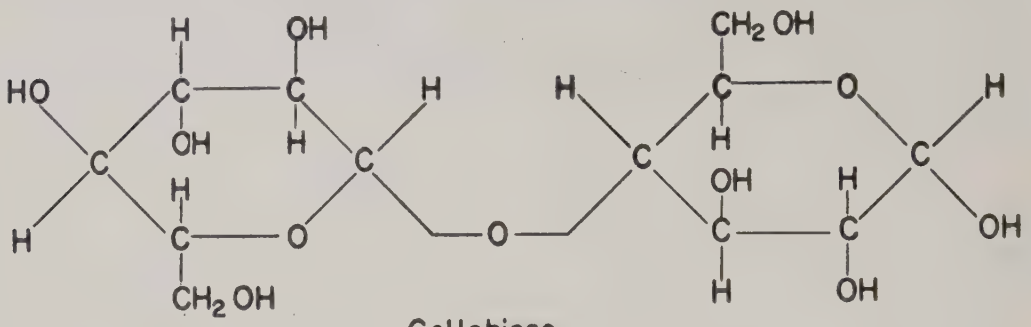
Reduced haemoglobin

Recent studies have shown that some amino acids survive in certain environments. ABELSON (1957) for example, has been able to identify free amino acids in calcareous fossil shells. The minute quantities of such residual amino acids, however, attest only to a successful entrapment mechanism, but do not nullify the conclusion that proteins and their amino acids are labile, are biologically decomposed, and probably contribute very little to the overall picture of petroleum genesis.

Carbohydrates

Carbohydrates account for a major portion of both plant and animal residues. They are classified as saccharides or disaccharides, both of which are water soluble, some tri- and tetrasaccharides, and finally as essentially insoluble polysaccharides such as starch, cellulose, and glycogen. Monosaccharides are characterized as either polyhydroxy aldehydes or ketones; the formation of polysaccharides

is accomplished by the elimination of water between the monomers to form oxygen bridges as shown below for the disaccharide, cellobiose:



Cellobiose

Carbohydrates are subject to a great variety of enzymatic reactions that degrade them into simpler substances. Under anaerobic conditions fermentation can occur in which a six-carbon monosaccharide, glucose, is converted into alcohol without the addition of oxygen. Other enzymatic processes are able to convert sugars into fat. As is well known, the cellulose

$$\underset{\text{glucose}}{C_6H_{12}O_6} \xrightarrow{\text{enzyme}} \underset{\text{ethyl alcohol}}{2C_2H_5OH} + 2CO_2$$

is bacteriologically ingested by herbivores depolymerized into mono- and di-saccharides that the animal can then metabolize as a source of energy. The enzymatic hydrolysis of carbohydrates is similar to the proteolytic degradation of proteins; in both cases the polymers are broken down into individual units or shorter chains with a minimum transfer of energy. The products of these reactions are subject to oxidation or reduction and are, therefore, sources of energy for life processes.

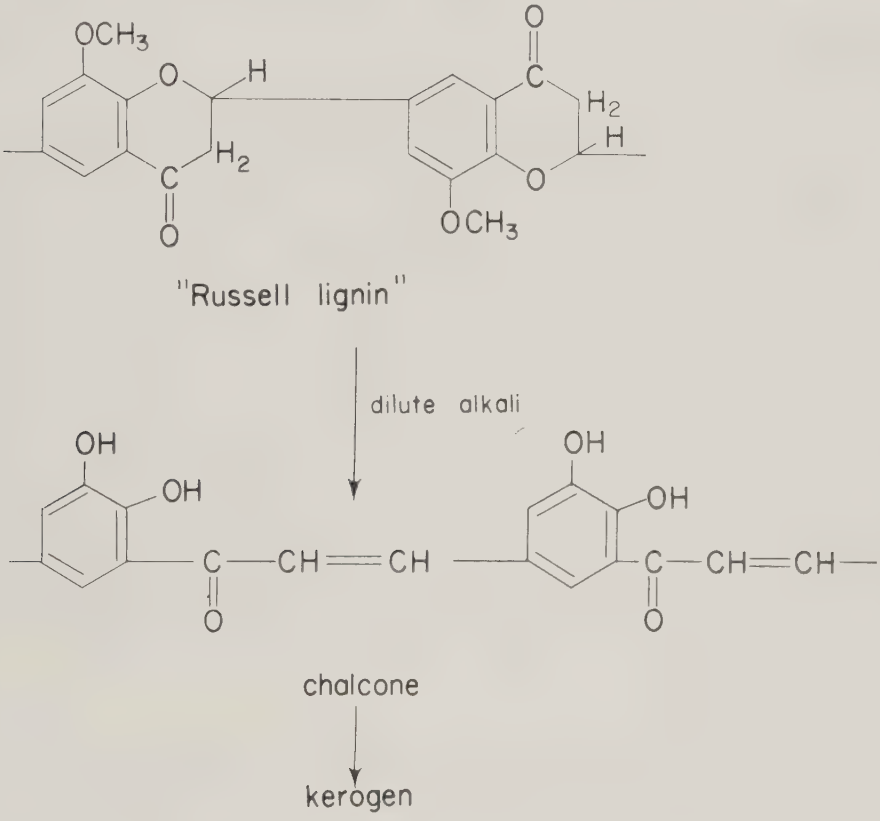
In general, the final products of carbohydrate metabolism are carbon dioxide and water. Although the widespread disappearance of appreciable residues of carbohydrates is evidence of their sensitivity to biochemical degradation, there is some evidence that under certain conditions carbohydrates can be protected and can, therefore, survive through geological time. Vallentyne and Whittaker (1956) have recently found free sugars in lake waters and sediments, and Theander (1952) reported the presence of galactose, glucose, mannose, arabinose, xylose and rhamnose in hydrolysates from sphagnum peat. The presence of small quantities of cellulose in the Eocene brown coals of Germany has also long been known. The very fact that a careful search for these residual minute quantities of sugars has been necessary is in itself significant in demonstrating the rapid and nearly total degradation of carbohydrates.

Lignin

Lignin, although not a normal constituent of marine organisms, can nevertheless be a major contributor to potential source beds for petroleum if it is carried into marine basins by rivers in the form of humic substances. Lignin has a

phenylpropane structure and is an aromatic compound. It also has a flavonoid structure which, when opened, develops acidic properties. Using the Russell formula for lignin (Russell, 1948) not as an actual representation of the structure but merely as a guide for illustration of potential degradative reactions, it is possible to demonstrate such rupture as shown below.

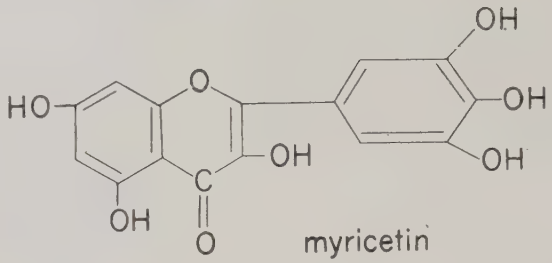
The product illustrated is demethylated because the methoxyl content of lignin is known to decrease rapidly on degradation. Double bonds may also be formed by the dehydration of β-hydroxy ketones, and the resultant derivative of catechol is extremely reactive towards condensation reactions. Ring opening with attendant formation of a chalcone, as shown below, has been postulated to occur rather rapidly in the presence of dilute (0·12 N) alkali (Fujise and Sasaki, 1938). Products of lignin degradation such as those shown must be considered in any conclusions regarding the fate of metabolic products in diagenetic environments. However, it is felt that these substances are the progenitors of kerogen and not precursors of petroleum.



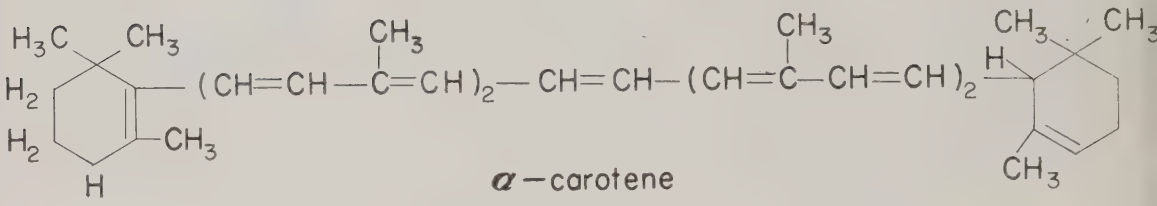
Pigments

Pigments of various types are produced by plants and animals and should be expected to contribute to sediments in which petroleum is formed. These pigments are numerous in nature and variety but for the sake of brevity, only

flavonoids, carotenes, and porphyrins will be considered. Examples of the first two types are shown below; the structure of protoporphyrin-III has already been given.



myricetin



α—carotene

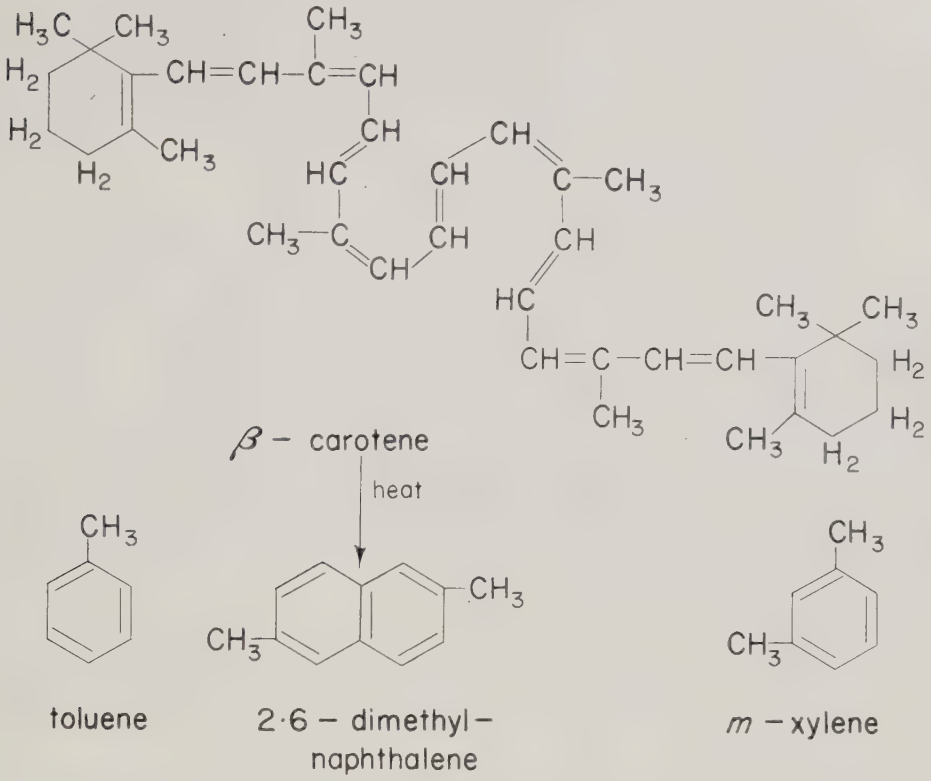
In general, the percentage of these pigments in biological substances are quite low and it may well be for this reason that flavonoids and carotenes have not been identified in marine sediments or crude oil even if they survive. Porphyrins, because of their high stability compared to other types of pigments, do survive and have been used successfully to demonstrate the low-temperature history of crude oil. Flavonoids, having the same basic structure as lignin, would be expected to hydrolyse to open-chain structures similar to those already discussed. Polyenes such as carotene, however, are extremely reactive and may well disappear by interaction with other substances or by intra- or intermolecular conversion. It may be noted in passing that β-carotene yields toluene, *m*-xylene, and 2:6-dimethylnaphthalene on heating at only 110°C. Degradation becomes understandable if the formula for β-carotene is written as shown on page 303 (Kuhn and Winterstein, 1932).

Considering that minute quantities of amino acids and proteins have been found as residues of major biological products, it seems likely that residues of these pigments, which represent only minor products, will eventually also be identified.

Lipids

To this point the discussion has covered proteins, carbohydrates, lignin and pigments; a consideration of lipids, which represents a third major biological product, has been left until last because of their particular importance.

Biochemically the term "lipid" refers to those substances that are insoluble in water but soluble in ether, benzene or chloroform. They may be simple lipids, such as esters of fatty acids and alcohols, or of fatty acids with glycerol; compound lipids containing nitrogen, phosphorus, or sulphur; or derived lipids such



Thermal degradation of β – carotene

as fatty acids, sterols or alcohols. For purposes of this discussion it is necessary to consider only the derived lipids inasmuch as many of the other varieties eventually break down to these simpler forms.

Natural glycerides, triesters of glycerol with various saturated and unsaturated fatty acids, disappear rapidly from depositional environments, presumably as a result of biological processes. Again, as in the case of proteins and polysaccharides, the glycerides are readily hydrolysable compounds yielding glycerol and fatty acids. The glycerol then serves as an energy source for biological processes, but the fate of the fatty acids, both saturated and unsaturated, is less well understood.

Fats and fatty acids are derived biologically from carbohydrates and amino acids. These reactions are dependent upon a two-carbon radical mechanism and, normally, give rise to even-numbered carbon chains. On degradation of a lipid, then there should be a preponderance of even-numbered fatty acids over those with odd numbers of carbon atoms in the chain. Saturated acids containing from four to twenty-six carbon atoms have been found in fats, but those with twelve, sixteen or eighteen carbon atoms are in the majority. Many unsaturated acids containing from one to six double bonds in unconjugated or conjugated positions

are also known (Cowan, 1953). A number of fatty acids, ranging from formic to nonanoic, have been isolated from crude oil (Hancock and Lochte, 1939).

The fate of fatty acids in a marine environment is not well understood. Rosenfeld (1948) found that unsaturation decreased with increase in depth of burial, but that bacterial dehydrogenation also was quite general. No clear-cut evidence for biological decarboxylation of fatty acids has been noted.

Chemical Conversion of Fatty Acids to Hydrocarbons

Inasmuch as fatty acids have basic hydrocarbon-like structures and apparently a relatively high degree of stability compared to other metabolic products, they present a class of substances of particular interest to the origin of petroleum. For this reason, it may be of some value to consider possible mechanisms by which such acids might be converted into the mixture of aliphatic, alicyclic, and aromatic hydrocarbons known to occur in crude oil. In any such consideration it is essential to recognize that biological degradation of metabolic products probably occurs very rapidly at the upper surface of uncompacted recent sediments. Following this initial decomposition, further reactions will most likely be chemical in nature. Because of the paucity of available information, much of what is to be suggested is of necessity speculative in nature.

The direct biochemical conversion of fatty acids to hydrocarbons in oil-forming quantities has not yet been reported. For this reason it is of considerable interest to note a number of potentially important enzymatically catalysed conversions that are known to occur in fatty acids and which may serve as important intermediate steps in petroleum synthesis.

Drawing an analogy from oxidative processes known to occur in the human liver, fatty acids might be converted by bacterial mechanisms into the following acids (Harper, 1957, pp. 196–198).

$RCH{=}CHCOOH$	α:β-unsaturated acid
$RCHOHCH_2COOH$	β-hydroxy acid
$RCOCH_2COOH$	β-keto acid

Human physiological reactions may also lead to the formation of keto or hydroxy acids, or acetone (Harper, 1957, pp. 196–198):

CH_3COCH_2COOH	acetoacetic acid
$CH_3CHOHCH_2COOH$	β-hydroxybutyric acid
CH_3COCH_3	acetone

These compounds, which represent intermediates in the oxidative degradation of fatty acids, are all more reactive compounds that the initial fatty acids and therefore represent substances which could enter into secondary reactions. Of particular interest in the potential ketonic cleavage of the β-keto acid in either weakly acidic or alkaline medium:

$$\underset{\beta\text{-keto acid}}{RCOCH_2COOH} \longrightarrow \underset{\text{ketone}}{RCOCH_3} + CO_2$$

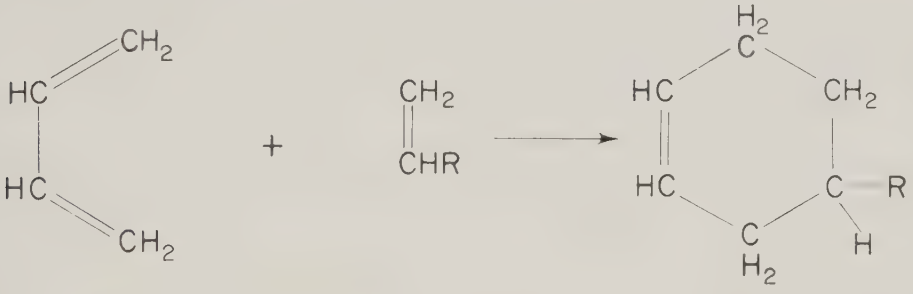
This reaction could lead to decarboxylation of a fatty acid and its conversion to an active ketone containing less oxygen and, moreover, an odd number of carbon atoms, assuming the initial acid to have had the usual even-numbered chain. An additional example of decarboxylation is that known to occur anaerobically through the medium of bacterial decarboxylase:

$$\underset{\text{pyruvic acid}}{2CH_3COCOOH} + H_2O \rightarrow \underset{\text{acetic acid}}{CH_3COOH} + CO_2 + \underset{\text{lactic acid}}{CH_3CHOHCOOH}$$

In this case dismutation occurs leading to the formation of the α-hydroxy acid, lactic acid (Porter, 1946).

To summarize then, although fatty acids may not be directly decarboxylated by bacteria, a number of changes are considered possible all of which result in oxygenated or dehydrogenated acids more reactive chemically that the parent substances. In addition to these new products, we must still consider those unsaturated acids initially available from the decomposition of glycerides within the overall picture of potential reactions leading to hydrocarbons.

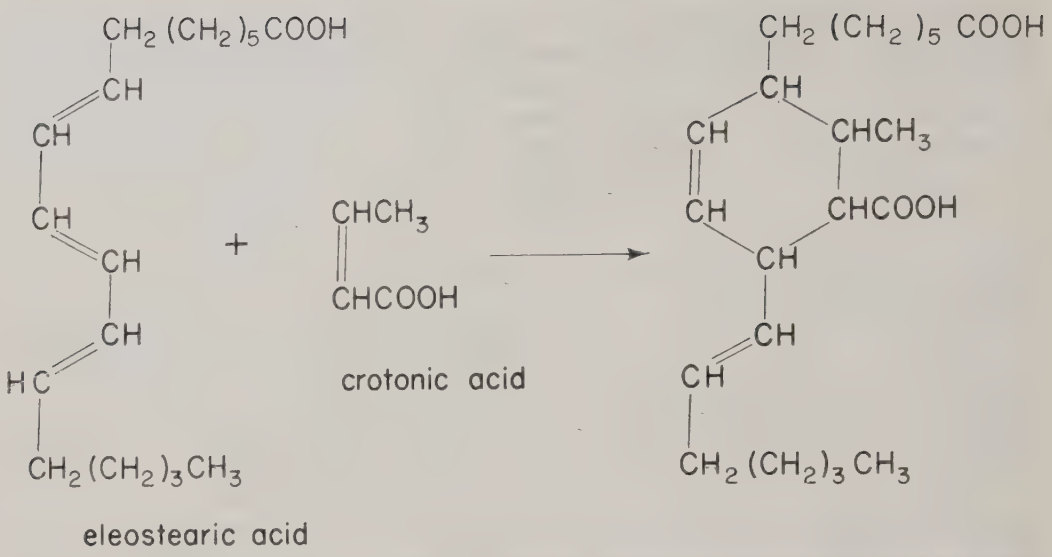
One of the most powerful mechanisms for the interaction of the above substances is the Diels–Alder reaction in which products may be formed from the following type of system:



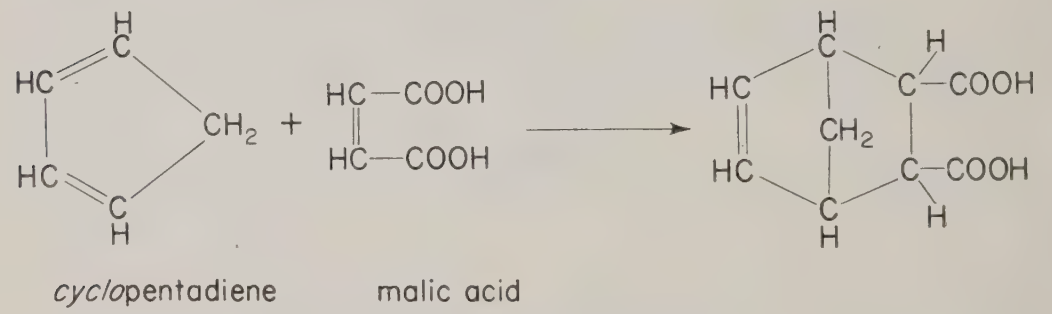
The R in this case usually represents an activating carbonyl (>C=O) group. As an example, the interaction between eleostearic acid from tung oil, and crotonic acid, would be expected to yield cyclic products. See top of page 306.

The Diels–Alder reaction is a purely chemical process operative at normal or elevated temperatures. This reaction can take place in hydrous media as shown by Sowa (1951), who found that α:β-unsaturated acids react readily with conjugated dienes at pH values below 7. See page 306.

Under a wide variety of conditions a broad spectrum of partially unsaturated ring compounds might be expected to result from the Diels–Alder reaction. Hydrogenation to alicyclic compounds or dehydrogenation to aromatic compounds is not inconceivable. There still remains, however, the problem of decarboxylating the products of this reaction to yield hydrocarbons. If the Diels–Alder reaction occurs rapidly in the zone of biological activity, then it is possible that the resultant acids are decomposed to ketones or aldehydes, in which case a mechanism for the reduction of the carbonyl group to a methylene (CH_2) or methyl (CH_3) group is necessary. No specific information on such a mechanism is available. Perhaps a more likely mechanism would be one in which the carbonyl



Example of a Diels–Alder reaction showing one of several possible products obtainable from eleostearic and crotonic acids.



group is reduced to an alcoholic hydroxyl which then splits out as water to leave as unsaturated bond:

$$RCH_2CH_2OH \rightarrow RCH{=}CH_2 + H_2O$$

The availability of hydrogen for saturation of this bond is probable in biologically active Recent sediments.

$$RCH{=}CH_2 + H_2 \rightarrow RCH_2CH_3$$

Many studies of the polymerization of drying oils by the Diels–Alder reaction have been reported. As an example, linoleic and linolenic acids, components of vegetable oils, have been found to dimerize by the Diels–Alder reaction following which unreacted double bonds isomerize into reactive, conjugated positions (Champetier and Petit, 1945).

Discussion

Although the preceding discussion is highly speculative, it does lead to a number of suggestions regarding the origin of petroleum.

(1) Current consideration that the complex organic constituents of marine muds and shales are converted to hydrocarbons by unknown processes and sources of energy is subject to revision. The contributions of proteins and carbohydrates to marine sediments are probably very small in view of the labile nature of these compounds; the major contribution to shales may actually be of terrestrial origin, namely humic substances derived from lignin. Such substances probably have little oil-forming potential thus explaining why some highly organic shales, such as the Chattanooga shale, are not major source beds.

(2) The most likely contributors to the formation of crude oil are the fatty acids and related substances. Initial biological degradation and associated Diels–Alder reactions may convert such substances into straight-chain compounds with both odd and even numbers of carbon atoms, to alicyclic compounds, and even, upon dehydrogenation, into aromatic substances. The numerous possibilities inherent in interaction of diverse saturated and unsaturated compounds could easily lead to a complex mixture of structures such as is found in crude oil.

(3) Many biochemical and chemical reactions, such as those that have been noted, result in products that still contain oxygen in the form of carbonyl, hydroxyl or carboxyl groups. Degradation of carboxyl groups to keto or hydroxy acids may be followed by decarboxylation. In view of these considerations, it would seem more profitable to search for those biochemical or chemical mechanisms able to reduce carbonyl or hydroxyl groups rather than for those that decarboxylate. According to Beerstecher (1954), bacterial hydrogenation of ketones, dehydrations, and the reduction of double bonds are all common reactions, but no specific examples are cited.

(4) Occurence of small quantities of hydrocarbons in recent sediments may be related to the fact that bacteria are able to produce traces of such material as part of their protoplasm (Zobell, 1951). The predominance of odd-numbered hydrocarbons over those with even-numbered chains may indicate a metabolic process in which a β-keto acid has been decarboxylated and the resulting ketone has been reduced. In general, however, reports of bacterial production of hydrocarbons are difficult to evaluate and not too convincing as far as the origin of petroleum is concerned (Beerstecher, 1954).

(5) Small amounts of condensed aromatic hydrocarbons can be formed by thermal degradation of carotenoid pigments, but it is considered more plausible that aromatics are formed by Diels–Alder mechanisms. The thermal history of a sediment and its accompanying oil cannot, however, be disregarded, for the rates of chemical reactions double for every 10°C rise in temperature.

(6) Based on these considerations it seems most likely that crude oil is generated in sediments containing high concentrations of lipids. If this is so, then petroleum could be formed in marine, brackish, or fresh-water environments, and the variety of hydrocarbons produced would be to a great extent dependent upon the contributory lipids. In this respect perhaps the paraffinic character of old oils and the naphthenic and aromatic character of young oils are related to variations in saturated and unsaturated lipids.

Numerous degradation products other than those mentioned in this paper have at one time or another been suggested as intermediates. Although some

intermediates may merit consideration, in general it is felt that those structures discussed above are by far the most important. Limitations in the scope of this paper have also prevented a discussion of sources of energy, such as heat and pressure, or of catalytic effects that may have influenced the course and speed of the reaction.

Acknowledgment—Many thanks are due to A. M. Pommer of the U.S. Geological Survey for his helpful comments and discussions. This report concerns work done by the U.S. Geological Survey on behalf of the Division of Research, U.S. Atomic Energy Commission.

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A hypothesis concerning the accumulation of sediment hydrocarbons to form crude oil

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Abstract—Small amounts of hydrocarbons, similar to those present in petroleum, are known to be dispersed throughout Recent marine sediments. However, their importance as major constituents of crude oils, if present in a like manner in ancient sediments at the time of deposition, is still being debated. Only recently has the significance of the Recent sediment hydrocarbons been indicated by the demonstration that crude oils and the sediments being deposited today contain certain identical kinds of hydrocarbons.

From an awareness of this close relationship and from a knowledge of the distributional differences which exist between Recent sediment and crude oil hydrocarbons, the process of forming oil appears mainly to be a physical one. Consequently, no extensive chemical alterations would be required by this hypothesis.

A study of hydrocarbon solubility in dilute colloidal electrolyte solutions suggested the nature of the physical process by which sediment hydrocarbons may be selectively removed from the source sediments and deposited in the correct proportions to form crude oil. The process appears to involve the solution, migration and release of hydrocarbons in waters containing natural solubilizers.

The similarity between the relative abundance of certain hydrocarbons in crude oil and the relative solubility of the same hydrocarbons in colloidal electrolyte solutions provides presumptive evidence that crude oils consist of Recent sediment hydrocarbons that were once solubilized in formation waters.

Introduction

One of the more intriguing problems in petroleum geology involves the mechanism of oil formation. Workers in this field tacitly assume that the origin and migration of oil are sequential processes in the sense that oil must originate before any can migrate. Consistent with this statement, it is generally believed that at least some oil originates in certain fine-grained sediments, which because of their above-average hydrocarbon content are designated as source rocks. Subsequently, the oil may, or may not, be expelled into other more porous beds.

With regard to the essential role of source rocks, namely the generation of oil, there is still no general agreement as to the nature of the source materials nor to the reactions which might transform them into petroleum. With regard to the migration of oil from source to reservoir rock, it has not been established whether oil moves in the form of a finely-dispersed emulsion or as a continuous oil phase, if indeed, the fine-grained sediments are permeable at all to oil in geological environments.

As Walters (1948) observed, deductions about source rocks have been based more on the nature of oil occurrence, itself, than on the chemical characteristics of the alleged source beds. The present understanding of this problem becomes even less certain when one points out, as did Hanson (1959), that only rarely can a petroleum be identified unequivocally with its particular source bed.

Although it has been convenient to discuss the origin and migration of oil separately, most geologists agree that the two processes are related. The purpose of this paper is to examine a very intimate relationship; namely, the suggestion by Baker (1959) that oil originates as a result of the migration of oil-like hydrocarbons in aqueous solution. To be more specific, the proposal is that oil consists

of Recent sediment hydrocarbons that have undergone solution, migration and release from waters containing natural solubilizers.

In the light of the above hypothesis, the essential role of source beds would be to provide the hydrocarbon constituents of oil, not the oil, *per se*. Moreover the transfer of hydrocarbons out of the fine-grained sediments into the more porous beds would involve primarily the question of permeability to a single phase, i.e. water. The hydrocarbons dispersed in sediments would exist as oil only after having undergone this solution and release process. Unfortunately, the release of sediment hydrocarbons as oil need not occur only in the more porous beds. If some discharge occurred in fine-grained sediments, the latter might be known as good source rocks, whereas in reality they would only be fine-grained sediments containing migrated hydrocarbons. This difficulty has already been pointed out by Bray and Evans (1959).

Petroleum-type hydrocarbons in Recent sediments

A significant research result of the last decade was the demonstration by Smith (1954) that finite amounts of paraffinic, aromatic and naphthenic hydrocarbons (as well as certain N—O—S hydrocarbon derivatives) are widely dispersed throughout a variety of Recent marine sediments. This announcement supported Whitmore's earlier suggestion (1943) that oil formation may only involve the selective accumulation of hydrocarbons produced by living organisms. However, the importance of the naturally-occuring sediment hydrocarbons as major constituents of crude oil continues to be debated by several investigators. Recently Meinschein (1959) has shown that crude oils and the sediments being deposited today contain certain identical kinds of hydrocarbons. He has also demonstrated that a close structural relationship exists between certain hydrocarbons present in sediments and crude oils and some organic compounds which comprise the lipid fraction of living organisms. For example, the tetracyclic hydrocarbons present in sediments and in crude oils contain carbon and hydrogen atoms that are combined in a manner almost identical to their arrangements in sex hormones, bile acids, cholesterol, and plant sterols. It appears that a majority of the hydrocarbons in petroleum are derived from plant and animal lipids. The formation of petroleum-type hydrocarbons probably takes place within the living organism, as discussed by Whitmore (1943), as well as in the sediments after burial, as is apparently required by Erdman *et al.* (1958). Although little is known about the actual reactions involved in the formation of these hydrocarbons, it appears that a geologic time factor is not required. Nor does it appear that hydrocarbon formation need be restricted to particular groups of sediments that may later be in proximity to known crude-oil deposits.

Distributional differences between sediment and petroleum hydrocarbons

Recent sediments do not inherit hydrocarbons in the amounts that characterize petroleum in reservoirs. Moreover, important differences exist in the relative amounts of certain homologous hydrocarbons dispersed in sediments and those concentrated in crude oil. These distributional differences which exist are signifi-

cant because they provide clues for understanding the nature of the process by which the dispersed hydrocarbons could become collected to form crude oil.

For example, it has been observed by Evans *et al.* (1957) that the normal paraffin hydrocarbons dispersed in a variety of Recent marine sediments exhibit a strong preference for odd-number carbon atoms. In the majority of crude oils, on the other hand, there is no such periodicity. As illustrated in Fig. 1, in the carbon number range between C_{20} and C_{32} the relative amounts of the individual *n*-paraffins in crude oils decrease smoothly with increasing carbon number. Thus, to convert sediment hydrocarbons into petroleum, some process must occur which selectively removes the hydrocarbons from the source sediments and deposits them in the correct proportions to form crude oil. Since no chemical alterations

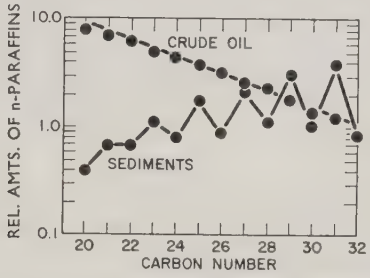

Fig. 1. Graph showing the distribution of *n*-paraffin hydrocarbons in Recent sediments and in crude oil.

would be required, the process need only be a physical one. But it is the process which results in oil formation; until it occurs, petroleum would not exist in sediments.

Distribution of hydrocarbons in crude oil

One might question why it was not necessary to label, or otherwise identify, the crude oil whose composition data are shown in Fig. 1. The reason is as Bray and Evans (1959) discussed; crude oils, in general, are found to contain heavy *n*-paraffins which are almost evenly distributed between molecules of odd and even numbers of carbon atoms. Although the total amount of *n*-paraffins in crude oils varies widely, the relative abundance or distribution of individual homologues is similar.

The fact that there is a similar distribution within a given family or class of hydrocarbons is a significant characteristic of the composition of crude oil. It is a fact that should be taken into account by any mechanism of oil formation. This result was first established by Forziati *et al.* (1944) from detailed studies of the molecular composition of the gasoline fractions of seven widely-different crude oils. These workers found that the naphtha fractions of different crudes are composed of the same hydrocarbons, the difference from one crude to another being essentially in the relative amounts of the classes of hydrocarbons (*n*-paraffins, *iso*paraffins, alkyl*cyclo*pentanes, alkyl*cyclo*hexanes, and aromatics) that are present. But most important, within each of the five classes, the individual hydrocarbons occur in nearly the same proportions in the different crudes. These observations were substantiated by Smith and Rall (1953) who investigated the

naphtha composition of twenty-one domestic and eleven foreign crude oils. The recent work of Martin and Winters (1959) also illustrates this important feature of crude oil composition.

As shown using Smith and Rall's data in Table 1, the individual hydrocarbons within a given class or family of hydrocarbons usually always occur in a definite proportion to each other. For example, methyl*cyclo*hexane is more abundant than *cyclo*hexane in crude oil. Although the ratio ranges between 1·09 and 3·13 in the thirty-two oils considered, the coefficient of variation is only 19·0 per cent. Likewise, within a given series of isomers, such as the C_6, C_7, and C_8 paraffins, the relative abundance generally decreases systematically with increased branching in the molecule.

Table 1. Relative abundance of hydrocarbons in crude oils according to Smith and Rall (1953).

Hydrocarbon type	Volume ratio of HC abundances in crude oils	
	21 domestic	11 foreign
Naphthenes methyl*cyclo*hexane to *cyclo*hexane	2·13	2·47
Aromatics toluene to benzene	4·27	4·78
Paraffins *n*-hexane to *n*-heptane	1·08	1·29

Although the finding of what Smith *et al.* (1959) term "order among chaos" has been extensively established with respect to the lower-boiling components of crude oil, it appears that the higher-boiling fractions of petroleum may also have a characteristic distribution of homologous hydrocarbons. This has been demonstrated as far as the heavier *n*-paraffins are concerned. That it may also be true for other classes of hydrocarbons is suggested by the work of Hood *et al.* (1959). From mass-spectrometric studies of the heavy distillate portions of petroleum, these workers have found that relatively few isomeric molecules make up the predominant portion of any hydrocarbon type at any given carbon number or boiling point. They find this result to be in agreement with the gasoline-range distribution of isomers reported by Forziati *et al.* (1944) and suggest, therefore, that a single picture of structural simplicity may apply to the entire distillate range of petroleum.

The fact that crude oils consist of only a relatively few of the theoretically-possible isomers may be a consequence of the finding by Meinschein (1959) that only a limited number of different hydrocarbons are initially present in the sediments.

The observation that a wide variety of crude oils appear to have a characteristic distribution of homologues and isomers is noteworthy since it implies the operation of a common, selective process of oil formation which would not greatly depend on the geographical and geological parameters. Although the whole composition of petroleum cannot be explained by thermodynamic considerations, it is possible that for a limited group of molecules having the same number of carbon atoms, i.e. a series of hydrocarbon isomers, thermodynamic equilibrium has actually been reached. Many workers such as Forziati and Rossini (1947) have used such reasoning and found good agreement between the characteristic distribution of particular isomers and the composition of their equilibrium mixture. However, the temperatures required ($>125°C$) to achieve such correspondence generally exceed those which were believed to have attained in the source sediments. Recently Baker (1959) has shown how this feature of crude oil composition may be a consequence of the solution and release of hydrocarbons from aqueous solutions containing natural solubilizers.

Migration in aqueous solution

Studies of hydrocarbon solubility in dilute organic acid salt solutions have suggested the nature of the physical process by which sediment hydrocarbons may be selectively collected to form crude oil. The process appears to involve sediment hydrocarbons which have dissolved in, moved with, and separated from the waters expelled from compacting sediments. The ability of this mechanism to account for the characteristic distribution of hydrocarbons in crude oil suggests that it may operate extensively in nature. A correspondence has been found between the relative abundance of hydrocarbons in crude oil and the relative solubility of the same hydrocarbons in the micelles formed by colloidal electrolytes. In other words, crude oils appear to consist of hydrocarbons that were once solubilized in formation waters.

When amphipathic molecules such as organic acid salts dissolve in water, they spontaneously associate to form clusters of individual molecules known as micelles. The presence of these colloidal particles in water increases the water solubility of hydrocarbons by providing hydrocarbon-like regions within the water in which hydrocarbons preferentially dissolve. Moreover, it is the solubility of hydrocarbons in just the micelles that is related to crude oil composition. Thus, Baker (1959) has found that the micellar solubility of toluene is between 2·6 and 5·7 times that of benzene. The observed micellar solubilities predict an excess of toluene over benzene that is typical of crude oils, as shown in Table 1. In a similar manner, it may be possible to explain why particular alkyl-substituted tetracyclic and higher polycyclic aromatics are more abundant than their unsubstituted parent hydrocarbons in crude oils, whereas in sediments the parent hydrocarbon is normally the more abundant homologue.

In an analogous manner, the characteristic distribution of the heavier *n*-paraffin hydrocarbons may also be explained. Since the micellar solubilities of the *n*-paraffin hydrocarbons show no periodicity, but decrease smoothly with increasing molecular weight, it is apparent from the solubility measurements reported by Baker (1958) that the formation waters would transport evenly

distributed, decreasing relative amounts of *n*-paraffin hydrocarbons such as characterize crude oils. All that would be required is that the sediments contain an excess of *n*-paraffin hydrocarbons over that needed to saturate the formation waters. The relative micellar solubilities of hydrocarbons are physicochemical facts, and all crude oils should reflect them. Hence, a characteristic or similar distribution of hydrocarbons within a given class of hydrocarbons would be expected by this hypothesis. Should the formation waters be undersaturated with respect to a particular group of hydrocarbons, then their distribution in crude oil would more nearly correspond to their distribution in the dispersed state, i.e. the sediments. Otherwise, the distribution of hydrocarbons in oil need not bear any relationship to their distribution in Recent sediments. In this connexion, Steven *et al.* (1956) have noted that the polycyclic aromatic fractions in crude oils are more complex than similar fractions obtained from a sediment. Such an increase in complexity is to be expected from the nature of the proposed hydrocarbon accumulation mechanism. As Meinschein (1959) has explained, "the waters in sediments would dissolve some of all the different hydrocarbons present in the sediments. The hydrocarbon solute of a water that has associated with a number of sediments would contain a more complex mixture of hydrocarbons than would be present in any one of these sediments."

Micellar solubilities and relative abundances of octane isomers

Studies of hydrocarbon solubilities in water and in dilute colloidal electrolyte solutions led to the following relation between crude oil composition and hydrocarbon solubility:

$$C_i = k(S_i - mS_i^\circ) \tag{1}$$

or

$$\frac{C_i}{S_i^\circ} = k\left(\frac{S_i}{S_i^\circ} - m\right) \tag{2}$$

where C_i is the volume ratio concentration of the ith hydrocarbon in crude oil, S_i and S_i° are the volume ratio solubilities of the ith hydrocarbon in the colloidal electrolyte solution and in pure water, respectively, and k and m are constants for a given petroleum and colloidal electrolyte solution. This equation states that the abundance of a hydrocarbon in crude oil is related to its micellar solubility ($m = 1$) and to the extent of subsequent leaching by water ($m > 1$), or to other processes that would *systematically* remove hydrocarbons *in proportion* to their water solubilities.

The solubilities of *n*-octane, 2:3-dimethylhexane and 2:2:4-trimethylpentane were measured in water (S_i°) and in water containing 0·1 weight per cent sodium naphthenate (S_i). The latter colloidal electrolyte solution was prepared by neutralizing the petroleum acids extracted from a Venezuelan crude oil with sodium hydroxide to a pH = 8·5. The micellar solubilities [$(S_i - mS_i^\circ)$, where $m = 1$] are not proportional to the relative abundances of these hydrocarbons in API Research Project 6 crude oil. However, by allowing for some water washing (that is, by selecting a value of m greater than unity in equation (1)), their abundances may be described by the above equations. As can be calculated with the aid of equation (1) using the values of C, S and S° for *n*-octane and 2:3-dimethyl-

hexane given in Table 2, the value of $m = 2{\cdot}70$ is such that 2:2:4-trimethylpentane (*iso*octane) would be completely removed from the micro-oil droplets that would form upon breakdown of the micelles and release of the heretofore-solubilized hydrocarbons. In other words, $m = 2{\cdot}70 > S_i/S_i^\circ = 1{\cdot}44$ for *iso*octane, and hence, according to equation (2), C_i for *iso*octane would be zero. This may explain why no *iso*octane has been detected in API Research Project 6 crude oil. During the migration of the oil toward the structurally-high parts of the reservoir rock, the oil is depleted (as by leaching by water) in the octane isomers in proportion to their water

Table 2. Solubility* of octane isomers in 0·1 weight per cent aqueous sodium naphthenate.

Hydrocarbon	C (vol. %)†	S° (p.p.m.)	S (p.p.m.)	(S/S°)
n-octane, C_i	1·90	0·090	0·311	3·46
2:3-dimethylhexane, C_j	0·07	0·200	0·550	2·75
2:2:4-trimethylpentane	—	0·900	1·296	1·44

* After ultrafiltration through Schleicher and Schuell Co. dense (average pore size = 150 mμ) membrane filters using the procedure described by Baker (1958).

† API Research Project 6 crude oil composition, according to Rossini and Mair (1959).

solubilities. Such a process, in the case of API oil, may have served to reduce the *iso*octane content to a level below which it can be detected.

Formation of crude oil

Formation waters would have the properties of micellar solutions if they contained small amounts of the water soluble salts of organic acids, such as the naphthenic acids present in petroleum. Although Kartsev *et al.* (1954) have reported the presence in certain oil field waters of as much as 5 g of sodium naphthenate per l., the presence of one-tenth this amount could result in the transport of significant amounts (5-10 p.p.m.) of hydrocarbons out of the compacting source sediments. It should be kept in mind, however, that in addition to organic acid salts, any organic molecule which can form association colloids in water could play a role in the migration process by enhancing hydrocarbon water solubility.

Since the solubilizing ability of micellar solutions is sensitive both to salt concentration and to dilution, hydrocarbons could be abruptly released from the waters as they pass from shale to sand and encounter environments of different ionic strengths. The break-up of micelles might also attend the diffusion of the micelles away from the shale-sand interface into the bulk of the water in the sand where they would, in effect, undergo dilution. The hydrocarbons which are released from micellar solution would appear for the first time as tiny oil droplets. They could coalesce and ultimately collect in the highest parts of the reservoir to form regions of high oil saturation.

This hypothesis appears to be consistent with the known geological facts about oil occurrence and the known chemical facts about the composition of sediment and crude oil hydrocarbons. However, it is not the writer's intention to

present the micellar solution mechanism of oil formation as an established fact. Instead, it is hoped that the ideas expressed herein will serve to provoke additional laboratory and field investigation. In the past variations in oil occurrence have been attributed to variations in source material and depositional environments or other geological descriptions that have of necessity been adjusted and redefined to fit the needs of the particular regions being investigated. In the future one might be able to understand oil occurrence as the manifestation of basic chemical and physical principles.

Summary

A summary of the proposed oil-forming mechanism is as follows:

(1) Recent sediments and crude oils contain some of the same kinds of hydrocarbons which appear to be derived from the organic compounds produced by living organisms.

(2) Since the majority of the hydrocarbons which may later become collected to form crude oil deposits appear to be present in the sediments relatively soon after deposition, no geologic time factor seems to be involved in hydrocarbon formation.

(3) Formation waters may contain natural solubilizers which would enhance the solubility of hydrocarbons. Such waters could selectively dissolve and release sediment hydrocarbons in proportions that correspond to hydrocarbon occurrence in crude oils.

(4) The solubilizing ability of the formation waters is thought to be sensitive both to salt concentration and to dilution. Consequently, hydrocarbons could be abruptly released as the interstitial waters pass from shale to sand or as the micelles diffuse throughout the sand.

(5) The hydrocarbons which are released would appear for the first time as tiny oil droplets (oil shows). They could coalesce and collect in the highest parts of the reservoir rock to form regions of high oil saturation (oil pools). The principles involved here are the classical ones of buoyancy and capillary pressure—entry pressure mechanics.

This hypothesis seems to be consistent with the known geological facts about oil occurrence and the known chemical facts about the composition of sediment and crude oil hydrocarbons.

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